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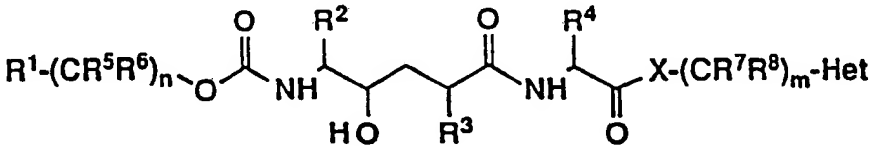

(I)

(57) Abstract

Compounds of formula (I) and pharmaceutically acceptable salts thereof and bioprecursors therefore wherein R^1 is C_1-C_6 alkyl, C_3-C_8 cycloalkyl, aryl, heterocyclyl or $CONR^9R^{10}$; R^2 is C_1-C_6 alkyl, C_3-C_8 cycloalkyl(C_1-C_4)alkyl, aryl(C_1-C_4)alkyl or heterocyclyl(C_1-C_4)alkyl; R^3 is C_1-C_6 alkyl, C_3-C_8 cycloalkyl, C_3-C_8 cycloalkyl(C_1-C_4)alkyl, aryl(C_1-C_4)alkyl, aryl(C_2-C_4)-alkenyl, heterocyclyl(C_1-C_4)alkyl or heterocyclyl(C_2-C_4)-alkenyl; R^4 is C_1-C_6 alkyl, C_3-C_8 cycloalkyl, aryl or heterocyclyl; each of R^5, R^6, R^7 and R^8 is independently H, C_1-C_6 alkyl or C_3-C_8 cycloalkyl; or R^5 and R^6, or R^7 and R^8 may be joined together to form a 3 to 8 membered carbocyclic ring; X is a 4-10 membered mono or bicyclic heterocyclic group containing carbon ring atoms and one ring nitrogen atom through which the group is attached to the adjacent carbonyl group; the group may be saturated or partially unsaturated and, in addition to the $-(CR^7R^8)_m$-Het substituent, it may be substituted by up to 4 further substituents independently chosen from F, C_1-C_6 alkyl, C_3-C_8 cycloalkyl, OR^{11} or NR^9R^{10}; Het is an imidazolyl or triazolyl group either of which may optionally be substituted by C_1-C_6 alkyl, C_3-C_7 cycloalkyl, NR^9R^{10} or $CONR^9R^{10}$, each of R^9 and R^{10} is independently H, C_1-C_6 alkyl or C_3-C_8 cycloalkyl, or R^9 and R^{10} may be joined together to form, with the nitrogen to which they are attached, a 4 to 8 membered nitrogen-containing heterocyclic group, R^{11} is H, C_1-C_6 alkyl or C_3-C_8 cycloalkyl; n and m are each independently 0, 1 or 2; wherein any alkyl or cycloalkyl group included in the aforementioned definitions may optionally be fully or partially substituted by fluorine; are inhibitors of retroviral proteases of utility in the treatment and prophylaxis of human retroviral infections.

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ANTIVIRAL PEPTIDES

The present invention relates to certain peptide derivatives containing a heterocyclic group which are useful in the treatment or prophylaxis of human retroviral infections.

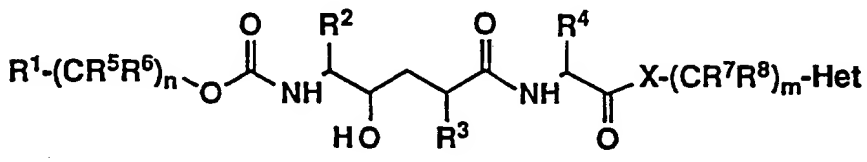
The human immunodeficiency virus (HIV), a retrovirus, is the causative agent of a variety of clinical conditions, the most serious of which are commonly termed AIDS (Acquired Immunodeficiency Syndrome), and ARC (AIDS-Related Complex). Infection with HIV is characterised by progressive breakdown of the immune system and CNS dysfunction. Severely immune deficient patients suffer from a wide range of opportunistic infections (e.g. pneumocystis carinii, human cytomegalovirus, or Candida), and cancers such as Kaposi's sarcoma. Loss of cells, particularly $CD4^+$ lymphocytes, following infection with HIV is an important factor in the progressive impairment of immune function. The infection of cells of monocyte/macrophage lineage with HIV also contributes to the observed pathology. Thus, successful infection of $CD4^+$ cells by HIV is a key step in the disease process.

HIV is a retrovirus; it encodes its genetic information in RNA, which is converted into DNA after the virus enters the host cell. An essential step in the retroviral replication cycle is the processing of an initial polypeptide precursor into mature structural and replicative proteins. This processing is carried out by a virus-coded protease and, in the absence of this enzyme activity, viral replication is blocked.

We have discovered that certain peptide derivatives linked to a heterocyclic group are potent inhibitors of retroviral proteases, both in a cell-free assay and in infected cells and, in addition, show antiviral activity in tissue culture assays. This

activity renders such compounds useful for the treatment and prophylaxis of retroviral infections, in particular, those caused by HIV.

Thus, the present invention provides compounds having the formula



(I)

and pharmaceutically acceptable salts thereof and bioprecursors therefor, wherein-:

R^1 is C_1-C_6 alkyl, C_3-C_8 cycloalkyl, aryl, heterocyclyl or $CONR^9R^{10}$;

R^2 is C_1-C_6 alkyl, C_3-C_8 cycloalkyl(C_1-C_4)alkyl, aryl(C_1-C_4)alkyl or heterocyclyl(C_1-C_4)alkyl;

R^3 is C_1-C_6 alkyl, C_3-C_8 cycloalkyl, C_3-C_8 cycloalkyl(C_1-C_4)alkyl, aryl(C_1-C_4)alkyl, aryl(C_2-C_4)-alkenyl, heterocyclyl(C_1-C_4)alkyl or heterocyclyl(C_2-C_4)-alkenyl;

R^4 is C_1-C_6 alkyl, C_3-C_8 cycloalkyl, aryl or heterocyclyl;

each of R^5, R^6, R^7 and R^8 is independently H, C_1-C_6 alkyl or C_3-C_8 cycloalkyl; or R^5 and R^6, or R^7 and R^8 may be joined together to form a 3 to 8 membered carbocyclic ring;

X is a 4-10 membered mono- or bicyclic heterocyclic group containing carbon ring atoms and one ring nitrogen atom through which the group is attached to the adjacent carbonyl group; the group may be

saturated or partially unsaturated and, in addition to the $-(CR^7R^8)_m$-Het substituent, it may be substituted by up to 4 further substituents each independently chosen from F, C_1-C_6 alkyl, C_3-C_8 cycloalkyl, OR^{11} or NR^9R^{10};

Het is an imidazolyl or triazolyl group either of which may optionally be substituted by C_1-C_6 alkyl, C_3-C_8 cycloalkyl, NR^9R^{10} or $CONR^9R^{10}$,

each of R^9 and R^{10} is independently H, C_1-C_6 alkyl or C_3-C_8 cycloalkyl, or R^9 and R^{10} may be joined together to form, with the nitrogen to which they are attached, a 4 to 8 membered nitrogen- containing heterocyclic group,

R^{11} is H, C_1-C_6 alkyl or C_3-C_8 cycloalkyl;

and n and m are each independently 0, 1 or 2;

wherein any alkyl or cycloalkyl group included in the aforementioned definitions may optionally be fully or partially substituted by fluorine.

In the above definition of R^1, R^2, R^3 and R^4, heterocyclyl means a 4 to 6 membered heterocyclic group containing as heteroatoms up to four nitrogen atoms, or an oxygen or sulphur atom optionally with one or two nitrogen atoms. The ring may be aromatic, or fully or partially saturated and may optionally be benzo-fused or substituted by C_1-C_6 alkyl, C_3-C_8 cycloalkyl, C_2-C_5 alkanoyl, C_1-C_4 alkoxy, halo, hydroxy, oxo or aryl. Preferred heterocyclyl groups are pyridyl, pyrimidinyl, thienyl, isoquinolyl and tetrazolyl.

In the above definitions of R^1, R^2, R^3 and R^4, aryl means phenyl optionally substituted with from 1 to 3 substituents each independently selected from C_1-C_6 alkyl, C_3-C_8 cycloalkyl, C_1-C_4 alkoxy, C_2-C_5 alkanoyl, hydroxy, halo, C_1-C_4 alkyl fully or partially substituted by fluorine, C_1-C_4 alkoxy fully or partially substituted by fluorine, phenyl, phenoxy, benzyl, benzoyl, phenylSO_2-, pyridyl, tetrazolyl, phenyltetrazolyl, NR^9R^{10} or $CONR^9R^{10}$; wherein R^9 and R^{10}

are as previously defined. Halo means fluoro, chloro, bromo or iodo.

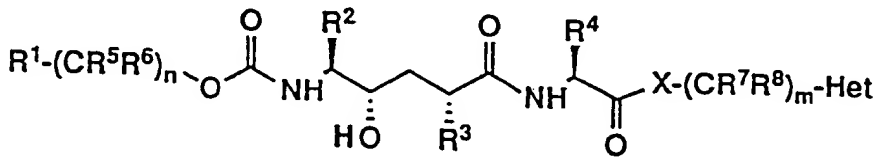
The imidazolyl or triazolyl group, Het, may be linked either by a ring carbon or ring nitrogen atom and may be unsubstituted or mono-, di or or tri-substituted. Triazolyl includes both 1,2,3 and 1,2,4-triazolyl groups.

Alkyl and alkoxy groups containing 3 or more carbon atoms may be branched or straight-chain. Any alkyl, alkoxy or cycloalkyl group included in the above definitions may optionally be fully or partially substituted by fluorine.

The term bioprecursor in the above definition means a pharmaceutically acceptable biologically degradable derivative of the compound of formula (I) which, upon administration to an animal or human being, is converted in the body to produce a compound of the formula (I). Examples include ester derivatives formed between the free hydroxy group in the compound of formula (I) and, for example, an amino acid (such as L-valine).

It will be appreciated that the compounds of formula (I) have a number of asymmetric carbon atoms and the invention includes all possible stereoisomers whether separated or not.

In one particular and preferred aspect of the invention there are provided compounds having the stereochemistry-:



(Ia)

In the above formula heavy bonds are used to indicate that the group lies above the plane of the molecule while a broken bond is used to indicate that the group lies below.

In the definition of R^1, aryl is preferably phenyl and heterocyclyl is preferably oxetan-3-yl or 1,1-dioxothietan-3-yl. R^1 is preferably t-butyl, isopropyl, oxetan-3-yl or 1,1-dioxothietan-3-yl and $(CR^5R^6)_n$ is absent; or R^1 is phenyl and $(CR^5R^6)_n$ is CH_2; or R^1 is H_2NCO-, CH_3NHCO- or $(CH_3)_2NCO-$ and $(CR^5R^6)_n$ is CH_2 or $CH(CH_3)$. Particularly preferred are compounds wherein R^1 is t-butyl, isopropyl or oxetan-3-yl and n is 0, most particularly where $R^1(CR^5R^6)_n-$ is t-butyl.

In the definition of R^2, aryl is preferably phenyl and heterocyclyl is for example pyridyl, pyrimidinyl or thienyl. R^2 is preferably aryl(C_1-C_4)alkyl; benzyl is particularly preferred.

In the definition of R^4, aryl is preferably phenyl and heterocyclyl is preferably pyridyl, pyrimidinyl or thienyl. R^4 is preferably C_1-C_6 alkyl; particularly preferred are isopropyl and sec-butyl (valine or isoleucine derivatives).

The heterocyclic group X is preferably a 4-6 membered saturated or monounsaturated group and is most preferably an azetidine, pyrrolidine, tetrahydropyridine or piperidine group; piperidine being particularly preferred.

R^7 and R^8 are preferably H and m is preferably 0 or 1.

In the definition of R^3, aryl is phenyl, unsubstituted or substituted as defined in the term aryl above, and heterocyclyl is for example pyridyl, pyrimidinyl, isoquinolyl or thienyl. R^3 is preferably aryl(C_1-C_4)alkyl or aryl(C_2-C_4)alkenyl; R^3 is most preferably benzyl optionally substituted in the phenyl

ring by fluoro, chloro, iodo, methyl, trifluoromethyl or trifluoromethoxy, or R^3 is 3-phenyl-prop-2-enyl or 3-phenylpropyl.

Particular and preferred individual compounds include:

1-[N-((R)-2-benzyl-(S)-5-(t-butoxycarbonylamino)-(S)-4-hydroxy-6-phenylhexanoyl)-(S)-valyl]-3-(imidazol-1-yl)azetidine,

1-[N-((R)-2-benzyl-(S)-5-(t-butoxycarbonylamino)-(S)-4-hydroxy-6-phenylhexanoyl)-(S)-valyl]-4-(imidazol-1-yl)piperidine,

1-[N-((S)-5-(t-butoxycarbonylamino)-(S)-4-hydroxy-6-phenyl-(R)-2-(3-phenylprop-2-en-1-yl)hexanoyl)-(S)-valyl]-4-(imidazol-1-yl)piperidine,

1-[N-((S)-5-(t-butoxycarbonylamino)-(S)-4-hydroxy-6-phenyl-(R)-2-(4-trifluoromethoxybenzyl)hexanoyl)-(S)-valyl]-4-(imidazol-1-yl)piperidine,

1-[N-((S)-5-(t-butoxycarbonylamino)-(R)-2-(4-chlorobenzyl)-(S)-4-hydroxy-6-phenylhexanoyl)-(S)-valyl]-3-(imidazol-1-yl)azetidine and

1-[N-((S)-5-(t-butoxycarbonylamino)-(S)-4-hydroxy-6-phenyl-(R)-2-(4-trifluoromethoxybenzyl)hexanoyl)-(S)-isoleucyl]-4-(imidazol-1-yl)piperidine.

In a second aspect of the present invention, there is provided a compound of formula (I), or a pharmaceutically acceptable salt thereof or bioprecursor therefor, for use as a medicament, especially for use in the treatment or prophylaxis of human retroviral infections, in particular HIV infections. The invention also includes the use of a compound of the formula (I), or of a pharmaceutically acceptable salt thereof or bioprecursor therefor, for the manufacture of a medicament for use in the prophylaxis or treatment of retroviral infections.

The invention further includes a pharmaceutical composition comprising a compound of the formula (I), or a pharmaceutically acceptable salt thereof or

bioprecursor therefor, and a pharmaceutically acceptable diluent or carrier.

The antiviral activity of the compounds of general formula (I) is established using in vitro assay systems. For example, the compounds of formula (I) are able to completely protect human T-cell line H9 for 7 days from the progressive effects of HIV infection. Untreated virus-infected cells display typical cytopathic effects such as formation of syncytia and cell death. In addition, virus particles produced from virus-infected cells treated with a compound of formula (I) are non-infectious.

Examples of infections which may be treated or prevented by the compounds of formula (I) include those caused by human or animal retroviruses, especially HIV-1. Clinical conditions which may therefore be treated or prevented include AIDS, ARC, and HIV related dementia. The compounds may also be used to block disease progression in symptomless infected individuals.

The compounds of formula (I) can be prepared using the coupling and protection techniques which are familiar to those skilled in the art of peptide chemistry.

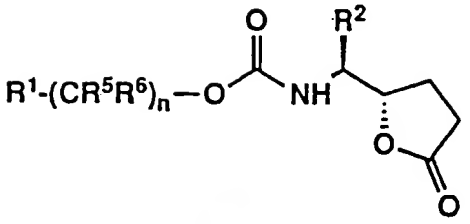
The process is illustrated, by way of example, by the routes shown below for the preparation of compounds of formula (Ia):

The procedure outlined in Scheme A, starts with a protected lactone (II). This is alkylated, using for example n-butyllithium or lithium hexamethyldisilazide followed by addition of a compound of formula R^3Br and separation of the desired isomer to give the product (III). The lactone ring is then opened by treatment with dilute alkali to give the corresponding hydroxy-acid and the hydroxy group is subsequently protected, for example as the t-butyldimethylsilyl derivative, by reaction with t-butyldimethylsilyl chloride in N,N-

dimethylformamide, followed by hydrolysis of the protected ester to give the intermediate (IV).

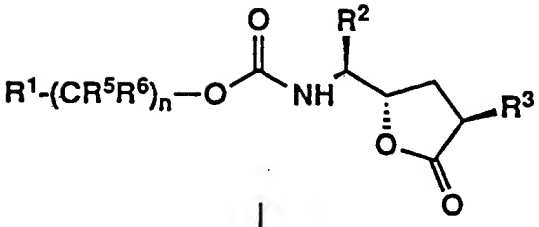
Coupling of this intermediate with the other fragment (VIII), to produce the compounds of formula (Ia), is illustrated in Scheme B. In this process, the protecting group on the intermediate (V) is removed (in the case of t-butoxycarbonyl by treatment with HCl) and the resulting compound (VI) is reacted with the N-protected amino acid Boc-NHCH(R^4)CO_2H, using an amide coupling reagent, to give the intermediate (VII). The N-protecting group from (VII) is then removed and the amine product (VIII) is coupled to intermediate (IV) to give intermediate (IX). The hydroxy protecting group X^1 is then removed from (IX) to give the final product of formula (Ia). In an alternative procedure the intermediate (IX) can be produced by coupling of the intermediate (X) with the intermediate (VI). The intermediate (X) can be prepared from intermediate (IV) by reaction with a carboxyl protected amino acid $H_2NCH(R^4)CO_2R$ and subsequent basic hydrolysis.

SCHEME A



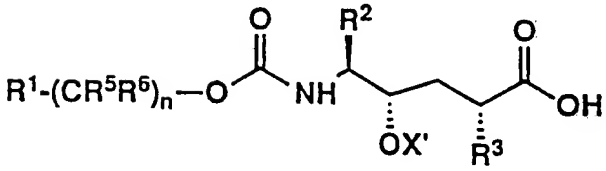
(II)

1. LiHMDS
2. R^3-Br



(III)

1. OH-
2. Protect alcohol and acid
3. Deprotect acid

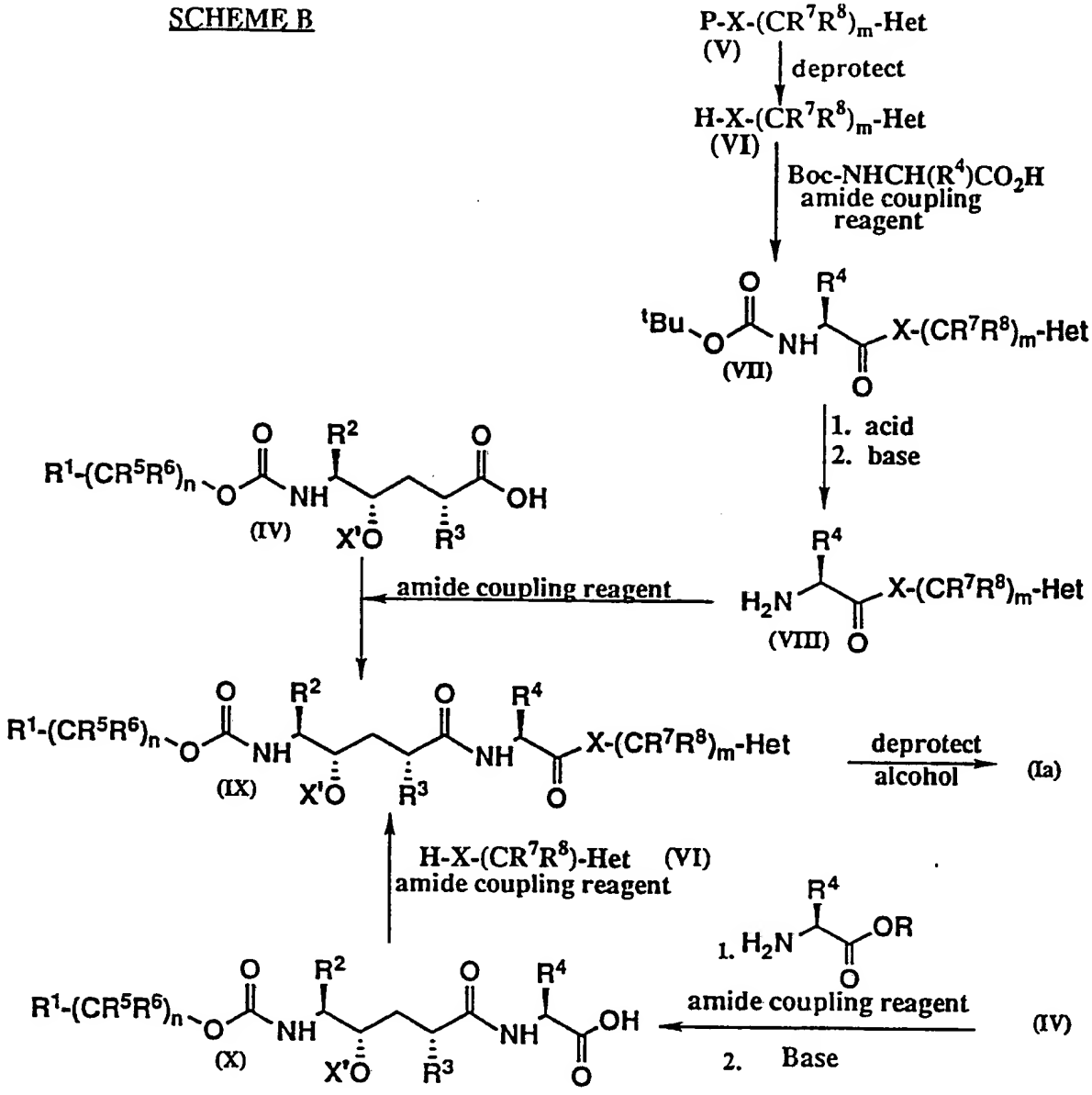


(IV)

Protecting group, X': tButyldimethylsilyl is preferred

R^1-$(CR^5R^6)_n$: t-Butyl or benzyl are preferred

SCHEME B



Amide coupling reagents:- e.g., 1-(3-dimethylaminopropyl)-3-ethylcarbodiimide

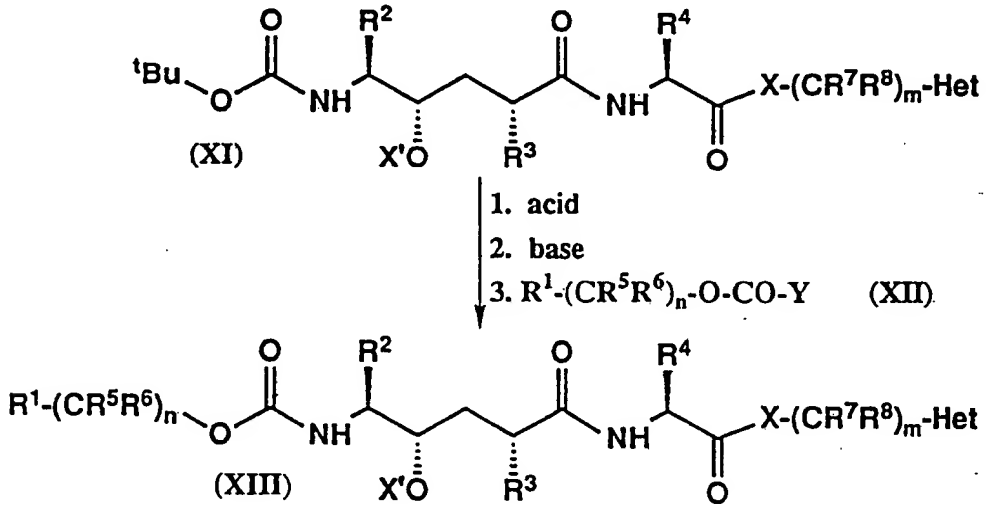
Protecting groups:- P; t-butoxycarbonyl or benzyloxycarbonyl are preferred

X'; t-butyldimethylsilyl is preferred.

R; methyl or ethyl are preferred

In another alternative procedure illustrated in Scheme C, removal of the t-butoxycarbonyl group from the intermediate (XI), by treatment with acid, for example trifluoroacetic acid, and subsequent reaction with a carbonate derivative of formula (XII) gives products of formula (XIII) in which the $R^1(CR^5R^6)_n$ group is other than t-butyl. For example, reaction of products of formula (XI) with 3-oxetanyloxy-carbonyloxysuccinimide gives the products (XIII) where R^1- is 3-oxetanyl and n is 0.

SCHEME C



X' is either H or a protecting group, *e.g.*, t-butyldimethylsilyl.

Y is a group which is susceptible to nucleophilic displacement - succinimidooxy is preferred.

The starting materials of formula (V) required for the procedure described above are in some cases literature compounds or they can be prepared by routine synthetic procedures from readily available starting materials. Thus for example reaction of 1(N-t-butoxycarbonyl)-3-hydroxy-azetidine with methanesulphonyl chloride followed by reaction with a compound of formula Het-H in the presence of a base gives the required compounds of formula (V) wherein X is an azetidine ring. Alternatively, for example, 1-(N-benzyloxycarbonyl)-4-ketopiperidine is converted to the 4-amino derivative by reaction with sodium cyanoborohydride in the presence of ammonium acetate. Subsequent reaction with dimethylformamide azine gives the 4-(1,2,4-triazol-4-yl) derivative.

Reaction of 1-(N-t-butoxycarbonyl)-4-keto-piperidine with the anion derived by reacting 1-diethoxymethyl)imidazole with n-butyllithium followed by treatment of the product with methanesulphonyl chloride in the presence of a base gives the N-protected-4-imidazol-2-yl(1,2,5,6-tetrahydropyridine) intermediate. Catalytic hydrogenation yields the corresponding N-protected-4-(imidazol-2-yl)-piperidine. Reaction of the above keto compound directly with a heterocyclic compound, e.g. imidazole, in the presence of thionyl chloride gives compounds of formula (V) wherein X is a tetrahydropyridine group, reduction again gives the piperidine derivative.

The intermediates of formula (V) in Scheme B where m does not equal 0 can be prepared by standard transformations from the appropriate precursors using, for example, nucleophilic opening of an epoxide group to introduce the Het group. Thus for example reaction of 1-(N-t-butoxycarbonyl)-4-ketopiperidine with trimethylsulphonium iodide in the presence of a base gives the 4-spiro-2'-oxirane. Reaction of this product with imidazole followed by elimination of the 4-hydroxy

group gives 1-(N-t-butoxycarbonyl)-4-(imidazol-1-yl)methyl-1,2,5,6-tetrahydropyridine; reduction gives the corresponding piperidine derivative.

The N-t-butoxycarbonyl (BOC-) derivatives of the naturally occurring amino acids used in the synthesis of the compounds of the formula (VII) are commercially available as are their hydroxysuccinimido esters. The corresponding intermediates derived from unnatural amino acids can be prepared by standard procedures (see, for example, M. J. O'Donnell et al, J. Amer. Chem. Soc., 1989, 111, 2353). The compounds of formula (II) can be prepared from the corresponding t-butyloxycarbonyl-protected amino-aldehydes (see D. H. Rich et al, J. Org. Chem., 1978, 43, 3624 and Y. Hamada et al, Chem. Pharm. Bull., 1982, 30(5), 1921) by reaction with ethyl propiolate (see A. H. Fray et al, J. Org. Chem., 1986, 51, 4828), followed by reduction to give the 5-t-butyloxycarbonylamino-4-hydroxy-6-phenylhexanoate. Cyclization by refluxing in toluene then affords the lactones of formula (II), as mixtures of diastereomers which can be separated by standard procedures.

In the above routes and specific Examples presented herein, certain hydroxy and amino-protecting and carboxy-activating groups are required. It will be apparent to those skilled in the art that the coupling and protection procedures described could be carried out by any standard method for peptide synthesis and these procedures are therefore included in the scope of the invention. The choice of a particular protecting group is dependent to a great extent upon the availability of the necessary reagent, its effect upon solubility of the protected compound, its ease of removal and the presence of other groups which might be affected by its use. For example, it is necessary in the above process to protect and deprotect particular amino groups in order to permit further reaction at the

regenerated amino group and the choice of protecting group for a given amino group will depend upon the role of said amino group in the overall reaction scheme. Amino protecting groups having varying levels of lability can be used. Such groups are known in the art and attention is directed to the reviews by Bodansky et al., "Peptide Synthesis", 2nd Ed., John Wiley & Sons, N.Y. (1976); Greene, "Protective Groups in Organic Synthesis", John Wiley & Sons, N.Y. (1981); McOmie, "Protective Groups in Organic Chemistry", Plenum Press, N.Y. (1973); and to Sheppard in "Comprehensive Organic Chemistry, The Synthesis and Reactions of Organic Compounds", Pergamon Press, N.Y. (1979), edited by E. Haslam, Part 23.6, pages 321-339.

Representative amino-protecting groups include, but are not limited to, aryloxycarbonyl such as benzyloxycarbonyl; substituted or unsubstituted aralkyl such as benzyl, trityl, benzhydryl and 4-nitrobenzyl; benzylidene; arylthio such as phenylthio, nitrophenylthio and trichloro- phenylthio; phosphoryl derivatives such as dimethylphosphoryl and 0,0-dibenzylphosphoryl; trialkylsilyl derivatives such as trimethylsilyl; and others as are described in U.S. Pat. No. 4,322,341. The preferred amino protecting group for use in the above sequence is t-butoxycarbonyl. Procedures for substituting said group on a given amino group are well known. In general they comprise acylating the appropriate amino compound with the corresponding carbonyl chloride or anhydride in a reaction inert solvent, e.g. water, methylene chloride or tetrahydrofuran, in the presence of a base (acid acceptor) e.g., sodium or potassium hydroxide when water is solvent; and, when an organic solvent is used, in the presence of a tertiary amine such as a triethylamine or pyridine. When an aqueous solvent system is used the pH of the reaction is typically held at about pH 8-10, and preferably at pH 9.

The protected amino groups are converted to the unprotected amino groups by procedures known to those skilled in the art as appropriate to the particular group employed. The t-butoxycarbonyl group is, for example, readily removed by treatment with dichloromethane saturated with hydrogen chloride gas.

Various hydroxy-protecting groups are also known and are described in the literative sources already cited above. A preferred hydroxy protecting group is t-butyldimethylsilyl. This is introduced as previously described and is readily removed by treatment with tetra-n-butylammonium fluoride in tetrahydrofuran at room temperature.

Activation of carboxy groups as a means of expediting a given acylation reaction is also methodology known to those skilled in the art. Especially useful in the herein described reaction sequence are the use of anhydrides and activated esters, particularly those esters derived from N-hydroxyphthalimide, N-hydroxysuccinimide or 1-hydroxybenzotriazole, all of which are used in peptide syntheses.

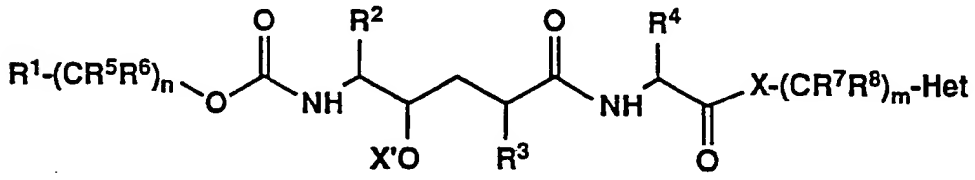
A dehydrative coupling agent is used to form the activated ester. Representative of such coupling agents are 1-cyclohexyl-3-(2-morpholinoethyl)-carbodiimide, N,N'dicyclohexylcarbodiimide, N,N'-carbonyldiimidazole, 1-(3-dimethylaminopropyl)-3-ethylcarbodiimide, ethoxyacetylene, diphenylketene and N-ethyl-5-phenylisoxazoline-3'-sulfonate. The reaction conditions for using such coupling agents are well described in the literature. In general they comprise the use of a reaction inert solvent and temperatures ranging from ambient to 100°C. The above-mentioned carbodiimide reagents are favoured since they permit use of ambient reaction temperature and afford satisfactory yields of the desired esters.

Upon completion of the coupling reactions leading

to the final products, the various protecting groups can be removed by the appropriate techniques previously discussed, and the compounds of the formula (I) isolated and purified using conventional procedures such as recrystallisation or column chromatography.

The above sequences can be adapted as appropriate to be performed with any of the variants claimed for R^1 to R^8, X and Het by appropriate selection of starting materials.

Thus, according to a further aspect, the invention includes a process for preparing a compound of the formula (I) which comprises removing the protecting group from a compound of the formula:



(IX)

wherein X^1 is a selectively removable hydroxy-protecting group, and isolating the compound of formula (I) and optionally forming a pharmaceutically acceptable salt thereof.

The preferred protecting group for X^1 is t-butyldimethylsilyl; this is removed by treatment with tetra-n-butylammonium fluoride in an organic solvent, preferably tetrahydrofuran.

The novel intermediates of formulae (VIII), (IX), and (X), also form part of this invention.

Examples of pharmaceutically acceptable salts of the compounds (I) are acid-addition salts, e.g.

sulfates, bisulfates, phosphates, lactates, mesylates, fumarates, citrates, succinates and gluconates.

In the treatment of patients having a retrovirus infection, especially HIV, the compounds (I) will be administered by any suitable route, e.g. by the oral, parenteral (e.g. subcutaneous, intravenous, intramuscular, or intradermal), rectal, nasal, topical (including buccal and sublingual) or vaginal routes. The formulations, which will contain an antiviral agent of the invention together with one or more pharmaceutically acceptable carriers and optionally other therapeutic agents, can be prepared according to conventional techniques well known in pharmacy. Oral dosage forms include in particular syrups, tablets and capsules which may contain flavouring agents in addition to an inert carrier. Tablets can be made by conventional compression or moulding techniques by compressing a powder of the appropriate ingredients, e.g. the antiviral agent in conjunction with a binder, diluent, lubricant and surface-active agent. Rectal formulations will be in suppository form, and vaginal formulations as, for example, tampons, creams or foams. Parenteral formulations will be in sterile form, e.g. as vials for injection containing aqueous or non-aqueous diluents, buffers and antioxidants so that the formulation will be isotonic with blood. In general, the appropriate dose of the anti-retroviral agents of the formula (I) will be from 1-50 mg/kg/day, preferably 1-25 mg/kg/day given in up to six divided doses per day. There may be of course instances where higher or lower dosages are merited according to the age, weight, degree of illness and response of the patient, and appropriate therapy will be as determined by the medical practitioner.

The compounds of formula (I) may be used in combination with other drugs, some of which may potentiate their activity. Such drugs include the

following:-

(a) Reverse transcriptase inhibitors such as AZT, ddI, ddC, foscarnet, TIBO compounds, dipyrido-diazepinones or 6-substituted acyclopyrimidine (HEPT) derivatives;

(b) gp120-CD4 blockers such as dextran sulphate and soluble CD4, including its combination with toxic agents such as pseudomonas toxin;

(c) tat antagonists, such as D-penicillamine;

(d) other retrovirals protease inhibitors, such as Ro 31-8959; and

(e) biological response modifiers including interferons, interleukins or colony stimulating factors, e.g. GM-CSF.

The compounds of the invention were evaluated for antiviral activity by dissolving the test compound in 50 µl of DMSO and diluting in RPMI 1640, a complex salts solution with a pH of 7.2, to 1 mg/ml. Testing was performed at 0.001, 0.01, 0.1, 1 and 10 µg/ml against HIV 1 (strain IIIB) in a human T-cell line (H9). Untreated control infections were initiated at the same time.

Seven days post infection, tissue culture supernatants were titrated for the presence of infectious virus on C8166 cells (human T-cell line). On days 3, 5, and 7 cultures were examined for appearance of syncytia. Control infections, untreated with drug, show typical viral cytopathic effects, including formation of syncytia and cell death. The IC_{100} quoted is the lowest test concentration affording complete protection to the culture. Using this test method, the compounds had an IC_{100} values in the range 0.1 to 10.0 µg/ml.

The preparation of certain starting materials and of the compounds of formula (I) will now be more particularly illustrated by reference to the following experimental Examples. The purity of compounds was

routinely monitored by thin layer chomatography using Merck Kieselgel 60 F_{254} plates. 1H-Nuclear magnetic reasonance spectra were recorded using either a Nicolet QE-300 or a Bruker AC-300 spectrometer and were in all cases consistent with the proposed structures. Chemical shifts are given in parts-per-million downfield from tetramethylsilane using conventional abbreviations for designation of major peaks: s, singlet; d, doublet; t, triplet; m, multiplet and b, broad. Optical rotations were read at a concentration of 0.1% in methanol at 25°C unless otherwise stated. All temperatures are in degrees celsius.

PREPARATION OF STARTING MATERIALS AND INTERMEDIATES

PREPARATION 1

(S)-5-[(S)-1-t-Butoxycarbonylamino-2-phenylethyl]-gamma-butyrolactone)

a) Ethyl (4S,5S)- and (4R,5S)-5-t-butoxycarbonylamino-4-hydroxy-6-phenylhex-2-ynoate

A solution of diisopropylamine (6.4 ml) in dry tetrahydrofuran (25 ml) was stirred under nitrogen at -25°C and a 1.6 molar solution of n-butyllithium in hexane (24.4 ml) was added over 5 minutes, keeping the temperature below -20°. After a further 15 minutes at -20° the solution was cooled to -70° and ethyl propiolate (3.8 g) was added dropwise over 10 minutes, keeping the temperature below -65°. The resulting yellow suspension was stirred at -70° for a further 20 minutes and then treated dropwise, over 10 minutes, with a solution of N-t-butoxycarbonyl-L-phenylalaninal (6.5 g, see J. R. Luly et al, J. Org. Chem., 1987, 52, 1487) in dry tetrahydrofuran (15 ml), again keeping the temperature below -65°. The clear yellow solution was stirred at -70° for 2 hours then treated with acetic acid (4 ml). The cooling bath was removed and the mixture was allowed to warm to -30° at which point water (100 ml) and ethyl acetate (100 ml) were added with vigorous stirring. Separation of the organic layer, followed by successive washing with 1 molar hydrochloric acid (50 ml), saturated aqueous sodium bicarbonate (50 ml) and saturated brine (50 ml), gave the crude product as an oil after drying (Na_2SO_4) and evaporation of the solvent. The oil was purified by silica gel chromatography using ethyl acetate-hexane (1:4) as the eluent. Evaporation of the product-containing fractions gave an oil which solidified on standing overnight. Recrystallisation from ether-hexane afforded the title compounds as an approximately 2:1 mixture (4S,5S:4R,5S) of diastereomers, (4.3 g), m.p. 98-99°. Found: C,65.62; H,7.42; N,4.33. $C_{19}H_{25}NO_5$

requires C,65.70; H,7.20; N,4.03%.
N.M.R. ($CDCl_3$) δ = 1.30-1.39 (m, 3H); 1.43 (s, 9H); 2.90-3.11 (m, 2H); 3.37-3.38 and 4.16-4.19 (2x brm, 1H); 3.93-4.04 (m, 1H); 4.22-4.33 (m, 2H); 4.51-4.56 (m, 1H); 4.77-4.79 and 4.87-4.90 (2 x brm, 1H); 7.24-7.35 (m, 5H).

b) Ethyl (4S,5S)- and (4R,5S)-5-t-butoxycarbonylamino-4-hydroxy-6-phenylhexanoate

The above product (1.17 g) was dissolved in ethanol (50 mg) and 5% Pd-$BaSO_4$ catalyst was added. The mixture was then hydrogenated at 50 psi (344.7 kPa) for 2 hours. Filtration, followed by evaporation of the solvent under vacuum, gave the title compounds (approximately 2:1 mixture of diastereomers) as a white solid, (1.18 g), m.p. 125-126°. Found: C,64.91; H,8.40; N,3.98, $C_{19}H_{29}NO_5$ requires C,64.95; H,8.26; N,3.98%.
N.M.R. ($CDCl_3$) δ = 1.24-1.33 (m, 3H); 1.39 and 1.42 (2 x s, 9H); 1.72-1.95 (m, 2H); 2.38-2.62 (m,2H); 2.77-2.98 (m,2H); 3.02-3.04 and 3.40-3.42 (2 x m, 1H exch. D_2O); 3.59-3.91 (m, 2H); 4.08-4.22 (m, 2H); 4.58-4.61 and 4.86-4.89 (2 x m, 1H); 7.22-7.37 (m, 5H).

c) (S)-5-[(S)-1-t-Butoxycarbonylamino-2-phenylethyl]-gamma-butyrolactone

The gamma-hydroxyester from (b) above was dissolved in 2.5% acetic acid-toluene (35 ml) and the solution was heated at reflux for 2 hours. After cooling and evaporation to dryness under vacuum, the residue was purified by silica gel chromatography, eluting with diethyl ether-hexane (40:60), to afford the title compound (0.4 g), m.p. 98-99°. Found: C,66.77; H,7.78; N,4.38. $C_{17}H_{23}NO_4$ requires: C,66.88; H,7.54; N,4.59%. m/e = 306 (MH^+).
N.M.R. ($CDCl_3$) δ = 1.42 (s, 9H); 2.11-2.19 (m, 2H); 2.51-2.58 (m, 2H); 2.87-3.02 (m, 2H); 4.00-4.07 (m,

1H); 4.47-4.52 (m, 1H); 4.63(d, J = 10, NH); 7.26-7.36 (m, 5H).

$[\alpha]_D^{25}$ -22.6° (c = 1, MeOH).

I. R. (KBr) 1775, 1690, 1525 cm^{-1}.

PREPARATION 2

(R)-2-Benzyl-(S)-5-t-butoxycarbonylamino-(S)-4-(t-butyldimethylsilyloxy)-6-phenylhexanoic acid

a) (R)-3-Benzyl-(S)-5-[(S)-1-t-butoxycarbonylamino-2-phenylethyl]-gamma-butyrolactone.

A cold (-10°) solution of hexamethyldisilazane (7.9 ml) in tetrahydrofuran (15 ml) was treated over 3 minutes with 1.6M n-butyllithium in hexane (23 ml), keeping the temperature below 0°. After a further 5 minutes at 0° the solution was cooled to -70° and a solution of (S)-5-[(S)-1-t-butoxycarbonylamino-2-phenylethyl]-gamma-butyrolactone (5 g) in tetrahydrofuran (38 ml) was added, keeping the temperature below -65°. The solution was stirred at -70° for 15 minutes before benzyl bromide (1.95 ml) in tetrahydrofuran (12.5 ml) was added over 1 minute and the solution stirred at -70° for an additional 10 minutes before being treated with acetic acid (6.5 ml) and the cooling bath removed. Water (50 ml) and ethyl acetate (50 ml) were added and the mixture allowed to warm to room temperature. Separation of the organic layer, followed by drying ($MgSO_4$) and evaporation of the solvent under vacuum gave the crude product as an oil. Chromatography on silica-gel, eluting with diethyl ether-hexane (50:50), gave the title compound as a clear oil (3.52 g). Found: C, 73.22; H, 7.50; N, 3.50. $C_{24}H_{29}NO_4$ requires C, 72.91; H, 7.34; N, 3.54%.

$[\alpha]_D^{25}$ -14° (c = 0.1%, MeOH)

N.M.R. ($CDCl_3$) δ = 1.37(s,9H); 1.95-2.30(m,2H); 2.79-3.20(m,5H); 3.92-4.01(m,1H); 4.21-4.25(m,1H); 4.52-

4.56(m,1H); 7.21-7.36(m,10H).

b) (R)-2-Benzyl-(S)-5-t-butoxycarbonylamino-(S)-4-(t-butyldimethylsilyloxy)-6-phenylhexanoic acid.

A suspension of the product from (a) (17.63 g) in dioxan (120 ml) and water (60 ml) was treated with sodium hydroxide (1N, 53.5 ml) at room temperature. The reaction was stirred for 3 hours before being acidified to pH 5 by the addition of acetic acid. After standing for a further 30 minutes the precipitate was filtered off and washed with water. This solid was dissolved in ethyl acetate, dried ($MgSO_4$) and evaporated under vacuum to a white solid which was triturated with hexane, filtered and dried to give the intermediate hydroxy acid (17.85 g). A solution of this hydroxy acid in N,N-dimethylformamide was treated with imidazole (29.38 g) and t-butyldimethylsilyl chloride (32.53 g) at room temperature. After stirring for 18 hours, the solvent was evaporated under vacuum, the residue treated with ice/water, 10% citric acid to pH 4, and extracted using 2 x 400 ml portions of ethyl acetate. The combined extracts were dried ($MgSO_4$) and evaporated under vacuum to a pale oil (29.2 g). A solution of this oil in tetrahydrofuran (240 ml) was treated with acetic acid (240 ml) and water (80 ml) at room temperature. After stirring for 2 hours at room temperature and 18 hours at 4°C the solution was evaporated under vacuum and the residue partitioned between water (400 ml) and ethyl acetate (400 ml). The separated organic layer was washed with water (2 x 400 ml), saturated brine (100 ml), dried ($MgSO_4$) and evaporated under vacuum to a pale oil. Chromatography on silica-gel, eluting with diethyl ether-hexane (70:30) gave the title compound as a white glass (22.4 g).

m/e 528 $(MH)^+$

N.M.R. (DMSO-d_6) δ = 0.10(s,6H); 0.95(s,9H); 1.30(m,10H); 1.35(m,1H); 1.95(m,1H); 2.40(m,1H); 2.72(m,2H); 2.85(m,1H); 3.60(m,1H); 3.75 (m,1H); 6.88(d,1H); 7.22(m,10H).

PREPARATION 3

5-Bromomethylisoquinoline

A solution of an isomeric mixture of 5- and 7-bromoisoquinoline (5:7, 40:60; 1.0 g; see Glyde and Taylor, J. Chem. Soc. Perkin Trans 2, 1975, 1783), in dry tetrahydrofuran (15 ml) was treated with 1.6M n-butyllithium in hexane (3.3 ml) at -70°C. The reaction was maintained at this temperature for 30 minutes and then a solution of dry dimethylformamide (0.74 ml) in dry tetrahydrofuran (5 ml) added. After a further 15 minutes, the reaction was quenched with ethanol (5 ml) and allowed to warm to room temperature. Saturated ammonium chloride solution (10 ml) and diethyl ether (15 ml) were then added sequentially and the organic phase separated, washed with saturated brine, dried ($MgSO_4$) and evaporated under vacuum. Purification by chromatography on silica gel eluting with hexane-ethyl acetate (50:50) and isolation of the higher running aldehyde gave 5-formylisoquinoline as an unstable yellow solid (0.05 g). Rf 0.3 (hexane-ethyl acetate 50:50).

A solution of this product (0.88 g) in dry methanol (20 ml) was treated with sodium borohydride (0.53 g) at 5°C and the resulting mixture allowed to warm to room temperature and maintained for 1 hour at this temperature. The solution was diluted with diethyl ether (20 ml) and water (10 ml) then added. The two phases were separated and the aqueous phases extracted with diethyl ether (20 ml). The two organic phases were combined, washed with saturated sodium chloride solution (20 ml), dried ($MgSO_4$), and evaporated under vacuum to give 5-hydroxymethyl-

isoquinoline as a pale yellow coloured powder, (0.37 g), m.p. 71-72°C. Found: C,75.06; H,5.72; N,8.66. $C_{10}H_9NO$ requires C,75.45; H,5.70; N,8.80%.

A solution of the above product (1.11 g) in glacial acetic acid (15 ml) was treated with 49% aqueous hydrobromic acid (30 ml) and the resulting mixture heated to reflux for 2 hours. The reaction was then concentrated under vacuum and the residue suspended in methylene chloride and basified with saturated aqueous sodium bicarbonate. The two phases were separated and the aqueous phase extracted with methylene chloride. The combined organic phases were washed with saturated aqueous sodium bicarbonate solution, dried ($MgSO_4$) for 0.5 hour and evaporated under vacuum at room temperature to give the title product as a colourless solid. The solid was azeotroped with toluene and then used directly (1.28 g).

N.M.R. ($CDCl_3$) δ = 4.90(s,2H); 7.50(t,1H); 7.70(d,1H); 7.90(m,2H); 8.65(d,1H); 9.25(s,1H).

PREPARATION 4

7-Bromomethylisoquinoline

7-Formylisoquinoline was obtained from the initial step of Preparation 3 as the lower running product and isolated as a yellow solid (0.08 g). Rf 0.25 (hexane-ethyl acetate 50:50). Reaction with sodium borohydride as described above gave 7-hydroxymethylisoquinoline, m.p. 129-130°C.

A solution of the hydrochloride salt of the above product (0.05 g) in thionyl bromide (0.5 ml) was heated to 60°C and maintained for 45 minutes. The reaction was then ice-cooled and excess water carefully added, followed by diethyl ether (15 ml). Concentrated aqueous ammonia was then added to pH 9 and the ether phase separated, washed with water and dried ($MgSO_4$). A solution of hydrogen chloride in isoproyl alcohol

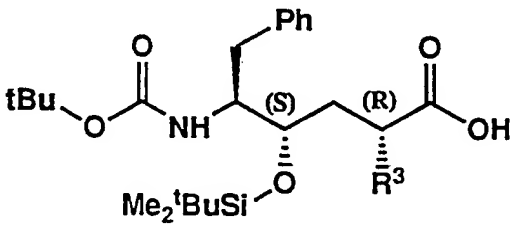
(0.06 ml, 5.9 N) was then added and the resulting cloudy suspension evaporated under vacuum at room temperature. The residue was azeotroped with toluene to give the product hydrochloride as a colourless solid (0.04 g).

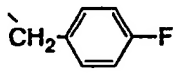
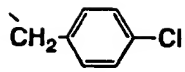
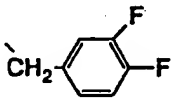
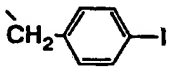
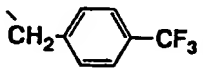
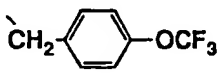
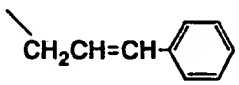
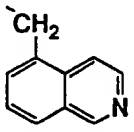
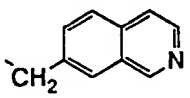
m/e $(MH)^+$ 222

N.M.R. (DMSO-d_6) δ = 5.05(s,2H); 8.10(d,1H); 8.25(d,1H); 8.30(d,1H); 8.45(s,1H); 8.65(d,1H); 9.70(s,1H).

PREPARATIONS 5-13

The following compounds of formula (IV) were prepared by the procedure described in Preparation 2 but using the appropriate substituted benzyl bromide or bromomethylisoquinoline to alkylate the gamma-butyrolactone in step (a) followed by ring opening and reaction with t-butyldimethylsilylchloride as described in step (b).



Prep No	R^3	MH^+	Analysis % C (Theoretical in brackets)	H	N
5		546.1	66.06 (66.02	8.34 8.31	2.51 2.56)
6		562.2			
7		564.6			
8		654.1	54.9 (55.1	6.75 6.78	2.07 2.14)
9		596	62.2 (62.5	7.71 7.44	2.26 2.35)
10		612.4	61.20 (60.90	7.30 7.30	2.30 2.30)
11		452	69.53 (69.40	8.59 8.55	2.52 2.53)
12		579	67.92 (68.26	7.92 8.02	4.72 4.82)[1]
13		579	67.77 (67.78	7.85 8.04	4.64 4.79)[2]

1. 0.1 mole H_2O 2. 0.33 mole H_2O

PREPARATION 14

1-(N-t-Butoxycarbonyl)-3-methanesulphonyloxy-azetidine

a) Azetidine-3-ol hydrochloride (2.10 g) was stirred in methylene chloride (40 ml) and diisopropylethylamine (2.59 g) was added followed by di-t-butyldicarbonate (4.36 g). The mixture was stirred at room temperature for 4 hours and the solvent was evaporated under vacuum. The residue was dissolved in ethyl acetate (200 ml) and washed with 1.5M hydrochloric acid (50 ml), saturated sodium bicarbonate solution (25 ml) and brine (25 ml). The organic layer was dried ($MgSO_4$), filtered and was evaporate under vacuum. Chromatography on silica gel, eluting with ethyl acetate-hexane (50:50), gave 1-(N-t-butoxycarbonyl)-3-hydroxy-azetidine as a white solid (2.57 g), m.p. 51-53°C. Found: C,55.29; H,8.70; N,7.98. $C_8H_{15}NO_3$ requires C,55.47; H,8.73; N,8.09%.

b) A solution of the above product (1.0 g) in methylene chloride (35 ml) was treated with methanesulphonyl chloride (0.75 ml) and pyridine (1.5 ml) and the mixture stirred for 3 days at room temperature. The solution was diluted with methylene chloride (75 ml), washed with aqueous citric acid solution (5%, 100 ml), saturated aqueous sodium bicarbonate (100 ml), dried ($MgSO_4$) and evaporated under vacuum to give the title product as a colourless oil (1.4 g). Found: C,43.00; H,6.80; N,5.50. $C_9H_{17}NO_5S$ requires C,43.03; H,6.77; N,5.58%. m/e 269 $(MNH_4)^+$. N.M.R. (DMSO-d_6) δ = 1.37(s,H); 3.24(s,3H); 3.88-3.96(m,2H); 4.17-4.28(m,2H); 5.25(m,1H).

PREPARATION 15

1-(N-Benzyloxycarbonyl)-(R)-3-methanesulphonyl-oxypyrrolidine

The title compound was prepared from 1-(N-benzyloxycarbonyl)-(R)-3-hydroxypyrrolidine (J.

Med. Chem., 1992, 35, 1764), using the procedure described in Preparation 14 above, except that triethylamine was used as base instead of pyridine, to give the product as an oil. Found: C,51.60; H,5.80; N,4.30. $C_{13}H_{17}NO_5S$ 1/6 H_2O requires C,51.63; H,5.78; N,4.63%. m/e MH^+ 300.

N.M.R. ($CDCl_3$) δ = 2.15(m,1H); 2.55(m,1H); 3.0(s,3H); 3.45-3.80(m,4H); 5.10(s,2H); 5.25(m,1H); 7.30(m,5H).

PREPARATION 16

1-(N-Benzyloxycarbonyl)-(S)-3-(paratoluene)sulphonyl-oxypyrrolidine

To a solution of 1-(N-benzyloxycarbonyl)-(R)-3-hydroxypyrrolidine (7.90 g) in dry tetrahydrofuran (100 ml) under nitrogen was added triphenylphosphine (13.58 g). The resulting solution was cooled to -30°C and methyl tosylate (10.06 g) added, followed by deithylazodicarboxylate (10.41 g) over a 0.5 hour period. After a further 1 hour, the reaction was allowed to warm to room temperature and maintained for 65 hours. The reaction was evaporated under vacuum, the residue dissolved in dichloromethane, washed with water, dried ($MgSO_4$) and evaporated under vacuum to an oil. Purification by chromatography on silica gel eluting with methylene chloride-methanol (98:2 to 96:4), followed by a second chromatography eluting with hexane-ethyl acetate (80:20 to 50:50) gave the product as a golden coloured oil (10.22 g). Found: C,60.59; H,5.68; N,3.67. $C_{19}H_{21}NO_5S$ requires C,60.78; H,5.64; N,3.73%.

$[\alpha]_D^{25}$ + 9° (c = 0.1%, MeOH)

N.M.R. ($CDCl_3$) δ = 1.85-2.25(m,2H); 2.45(s,3H); 3.40-3.65(m,4H); 4.95-5.15(m,3H); 7.20-7.35(m,7H); 7.75(d,2H).

PREPARATION 17

1-(N-t-Butoxycarbonyl)-4-methanesulphonyloxypiperidine

The title compound was prepared from 1-(N-t-butoxycarbonyl)-4-hydroxypiperidine, following the procedure described in Preparation 14, m.p. 85-86°C. Found: C,47.2; H,7.66; N,4.91. $C_{11}H_{21}NO_5S$ requires C,47.3; H,7.58; N,5.02%.

N.M.R. ($CDCl_3$) δ = 1.5(s,9H); 1.85(m,2H); 2.0(m,2H); 3.08(s,3H); 3.35(m,2H); 3.75(m,2H); 4.9(m,1H).

PREPARATION 18

1-(N-t-Butoxycarbonyl)-3-(imidazol-1-yl)azetidine

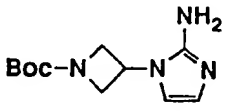
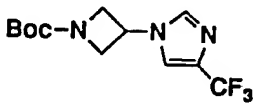
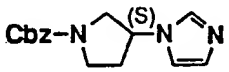
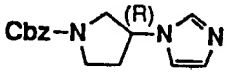
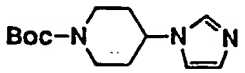
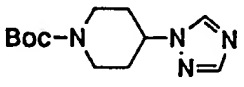
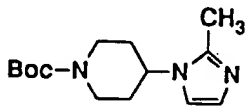
A solution of imidazole (0.41 g) in N,N-dimethylformamide (30 ml) was treated with sodium hydride (60%, 0.24 g) and the mixture stirred at room temperature for 1 hour. 1-(N-t-Butoxycarbonyl)-3-methanesulphonyloxy-azetidine (Preparation 14). (1.4 g) was added and the mixture was heated at 75°C for 3 days. The solvent was then removed under vacuum and the residue dissolved in ethyl acetate (50 ml), washed with water (2 x 50 ml), dried ($MgSO_4$) and evaporated under vacuum to give a colourless oil. Purification by chromatography on silica gel eluting with methylene chloride-methanol-concentrated aqueous ammonia (93:7:1) gave the title compound as an oil (0.72 g).

m/e 224 $(MH)^+$

N.M.R. (DMSO-d_6) δ =1.40 (s, 9H); 3.95-4.06 (m, 2H); 4.26-4.35 (m, 2H); 5.12 (m, 1H); 6.96 (s, 1H); 7.43 (s, 1H); 7.79 (s, 1H).

PREPARATIONS 19-25

The following compounds of formula (V) were prepared following the procedure described in Preparation 18 above but using the appropriate starting material from Preparations 14 to 17 and reacting with the appropriate substituted or unsubstituted imidazole or triazole.

Prep No	P-X-$(CR^7R^8)_m$-Het	$(MH)^+$	Analysis % (Theoretical in brackets) C	H	N
19		239.3	55.1 (55.4	7.50 7.67	22.9 23.5)
20			NMR: 1.33(s,9H) 4.05 (m,2H); 4.24(m,2H); 5.1(m,1H); 7.99(s,1H); 8.15(s,1H).		
21		272	65.57 (65.60	6.37 6.25	15.04 15.25)[1]
22		272	65.69 (65.60	6.33 6.25	15.25 15.25)[1]
23		252			
24		253			
25		265.9	61.0 (61.3	8.92 8.82	15.6 15.3)[2]

1. 0.05 mole CH_2Cl_2 2. Hemihydrate

PREPARATION 26

(a) 1-(N-t-Butoxycarbonyl)-4-(4-methylimidazol-1-yl)piperidine

Reaction of 4-methylimidazole with 1-(N-t-butoxycarbonyl)-4-methanesulphonyloxypiperidine gave two regioisomeric products, which were separated by chromatography on silica-gel, eluting with dichloromethane : methanol : concentrated aqueous ammonia (96:3.5:0.5). Major isomer, Rf 0.47. m/e 265.9 $(MH)^+$

N.M.R. (DMSO-d_6) δ = 1.43 (s, 9H); 1.7 (dq, 2H); 1.93 (bd, 2H); 2.07 (s, 3H); 2.83 (m, 2H); 4.05 (m, 3H); 6.95 (s, 1H); 7.55 (s, 1H).

(b) 1-(N-t-Butoxycarbonyl)-4-(5-methylimidazol-1-yl)piperidine

Minor isomer, Rf 0.52. m/e 265.9 $(MH)^+$

N.M.R. (DMSO-d_6) δ = 1.43 (s, 9H); 1.7 (dq, 2H); 1.9 (bd, 2H); 2.05 (s, 3H); 2.88 (m, 2H); 4.05 (m, 3H); 6.6 (s, 1H); 7.65 (s, 1H).

PREPARATION 27

1-(N-t-Butoxycarbonyl)-4-(imidazol-2-yl)-(1,2,5,6-tetrahydropyridine

(a) 1-(Diethoxymethyl)imidazole (6.8 g) was stirred in dry tetrahydrofuran (50 ml) under nitrogen at -40°C. n-Butyllithium (25 ml, 1.6N in hexane) was added at such a rate that the temperature remained below -35°C. 1-(N-t-Butoxycarbonyl)-4-keto-piperidine (2.65 g) in dry tetrahydrofuran (10 ml) was added dropwise over 10 minutes, keeping the temperature below -40°C, and the resulting mixture stirred at -40°C for 2 hours. The reaction mixture was stirred with hydrochloric acid (50 ml, 0.1N) for 15 minutes, ethyl acetate (50 ml) was then added and the resulting mixture stirred for 5 minutes. The organic layer was separated and the aqueous layer extracted with ethyl acetate (1 x 50 ml).

The combined organic extracts were washed with saturated sodium bicarbonate solution (1 x 50 ml), then with saturated sodium chloride solution. The organic layer was then dried ($MgSO_4$) and evaporated to a yellow oil, 8.0 g. Chromatography on silica-gel, eluting with ethyl acetate: methanol: concentrated aqueous ammonia (90:10:1), yielded 1-(N-t-butoxycarbonyl)-4-hydroxy-4-imidazol-2-ylpiperidine a cream solid, 2.2 g. m/e 268.0 $(MH)^+$.

(b) The above product (267 mg) was stirred with diisopropylamine (350 ml) in dry dimethylformamide (2 ml) at 0°C. Methanesulphonyl chloride (155 ml) was added in one portion, and the reaction mixture stirred for 2 hours at 0°C. Further portions of diisopropylamine (350 ml) and methanesulphonyl chloride (155 ml) were then added and the reaction mixture stirred for 16 hours at room temperature. The resulting mixture was diluted with water (4 ml), adjusted to pH 9 with 1M sodium hydroxide solution, and extracted with ethyl acetate (3 x 10 ml). The combined organic extract was dried ($MgSO_4$), and evaporated to a gum, 260 mg. Chromatography on silica-gel, eluting with dichloromethane: methanol: concentrated aqueous ammonia (95:5:1) gave the title product as a yellow gum, 144 mg. m/e 250.1 $(MH)^+$.

N.M.R. ($CDCl_3$) δ = 1.46 (s, 9H); 2.6 (bs, 2H); 3.53 (t, 2H); 4.0 (bs, 2H); 6.3 (m, 1H); 7.0 (s, 2H); 9.4 (bs, 1H).

PREPARATION 28

1-(N-t-Butoxycarbonyl)-4-imidazol-2-ylpiperidine

The product from Intermediate Preparation (27) (0.55 g) was dissolved in ethanol (30 ml) and hydrogentated at 30 p.s.i. (2.0 bar) with palladium on carbon catalyst (200 mg, 10%). Filtration of catalyst and removal of solvent gave a foam, 0.55 g. m/e 252.1 $(MH)^+$.

N.M.R. ($CDCl_3$) δ = 1.4 (s, 9H); 1.68 (qd, 2H); 1.95 (bd, 2H); 2.75 (bt, 2H); 2.92 (bt, 1H); 4.1 (bd, 2H); 6.9 (s, 2H); 8.77 (bs, 1H).

PREPARATION 29

1-(N-t-Butoxycarbonyl)-4-(imidazol-1-yl)methyl-1,2,5,6-tetrahydropyridine

(a) A 60% oil dispersion of sodium hydride (4 g) in dry dimethylsulphoxide (100 ml) was washed free of oil with hexane and then heated at 70°C with stirring for 1 hour. Dry tetrahydrofuran (100 ml) was added and the reaction cooled to -20°C. A solution of trimethylsulphonium iodide (20.4 g) in dimethylsulphoxide (80 ml) was then added, followed by 1-(N-t-butoxycarbonyl)-4-ketopiperidine (19.9 g) in dry tetrahydrofuran (100 ml) and the reaction stirred for 0.5 hour at -10°C and then 1 hour at room temperature. Water (500 ml) was then added and the mixture extracted with ethyl acetate (3 x 250 ml). The combined extracts were then washed with brine, dried ($MgSO_4$) and evaporated under vacuum. Purification by chromatography on silica gel eluting with cyclohexane-ether-isopropyl alcohol (60:40:1) gave 1-(N-t-butoxycarbonyl)-piperidine-4-spiro-2'-oxirane as a colourless solid, (19.6 g), m.p. 65-66°C. Found C, 62.11; H, 9.06; N, 6.55. $C_{11}H_{19}NO_3$ requires C, 61.97; H, 8.92; N, 6.57%.

(b) A stirred solution of imidazole (1.91 g) in dry acetonitrile (30 ml) under nitrogen was treated with an 80% oil dispersion of sodium hydride (0.84 g) and the resulting mixture heated to 60°C until solution occurred. After 15 minutes the product from step (a) (2.0 g) was added and the reaction maintained for 5 hours. After allowing the reaction to stand at room temperature overnight, the solvent was evaporated under vacuum and the oily residue partitioned between

methylene chloride (40 ml) and water (20 ml). The organic phase was separated and washed with water, dried ($MgSO_4$) and evaporated under vacuum. Purification by chromatography on silica gel, eluting with methylene chloride-methanol-concentrated 880 aqueous ammonia (95:4:1) gave 1-(N-t-butoxycarbonyl)-4-hydroxy-4-(imidazol-1-yl)methylpiperidine as a colourless powder (2.13 g). Found C, 58.35; H, 8.30; N, 14.52. $C_{13}H_{23}N_3O_3$ $^1/_{10}$ CH_2Cl_2 requires C, 58.43; H, 8.07; N, 14.50%.
m/e 282 $(MH)^+$.

(c) A stirred solution of the product from step (b) (2.0 g) and triethylamine (5.44 ml) in dry methylene chloride (80 ml) at 0 to 5°C was treated with a solution of methanesulphonyl chloride (2.20 ml) in dry methylene chloride (10 ml) and the resulting mixture allowed to warm to room temperature and maintained for 14 hours. The reaction mixture was then washed with water, dried ($MgSO_4$) and evaporated under vacuum. Purification of the residue by chromatography on silica gel eluting with methylene chloride-methanol-concentrated 880 aqueous ammonia (96:4:0 to 95:4:1) gave the product as a golden coloured oil (1.36 g). Found C, 59.93; H, 7.60; N, 14.79. $C_{13}H_{21}N_3O_2$ $^1/_4$ CH_2Cl_2 requires C, 60.14; H, 7.61; N, 14.77%.
m/e $(MH)^+$ 264
N.M.R. ($CDCl_3$) δ = 1.45 (s, 9H); 1.97 (m, 2H); 3.50 (t, 2H); 3.90 (s, 2H); 4.50 (s, 2H); 5.50 (s, 1H); 6.90 (s, 1H); 7.10 (s, 1H); 7.55 (s, 1H).

PREPARATION 30

1-(N-t-Butoxycarbonyl)-4-(imidazol-1-yl)methyl-piperidine

A solution of the product from Preparation 29 (1.33 g) in absolute ethanol (25 ml) was hydrogenated with stirring over 10% palladium on charcoal (0.3 g) at 50 p.s.i. (3.5 bar), room temperature, for 4 hours. The

reaction mixture was then filtered and evaporated under vacuum, azeotroping with methylene chloride. Purification by chromatography on silica gel eluting with methylene chloride-methanol (96:4) gave the product as a colourless oil (1.09 g). Found C, 63.08; H, 8.60; N, 15.50. $C_{13}H_{23}N_3O_2$ 1/4 CH_2Cl_2 requires C, 63.36; H, 8.74; N, 15.84%. m/e $(MH)^+$ 266
N.M.R. ($CDCl_3$) δ = 1.15 (m, 2H); 1.45 (s, 9H); 1.58 (m, 2H); 1.85 (m, 1H); 2.65 (t, 2H); 3.80 (d, 2H); 4.10 (m, 2H); 6.90 (s, 1H); 7.10 (s, 1H); 7.45 (s, 1H).

PREPARATION 31

1-(N-t-Butoxycarbonyl)-4-(imidazol-1-yl)-1,2,5,6-tetrahydropyridine

Imidazole (8.20 g) was stirred in dry methylene chloride (30 ml) at -10°C and thionyl chloride (4.8 ml) in dry methylene chloride (30 ml) added. The resulting slurry was allowed to warm to room temperature and after 2 hours a solution of 1-(N-t-butoxycarbonyl)-4-keto-piperidine (6.0 g) in dry methylene chloride (50 ml) was added dropwise. The reaction mixture was stirred overnight and then evaporated under vacuum. Potassium carbonate (6.0 g) in water (30 ml) was added to the oily residue and the product extracted with methylene chloride (2 x 60 ml). The combined extracts were washed with water (40 ml), dried ($MgSO_4$) and evaporated under vacuum. Purification by chromatography on silica gel eluting with ethyl acetate-methanol (100:0 to 90:10), gave the product as an oil (1.0 g).
N.M.R. ($CDCl_3$) δ = 1.50 (s, 9H); 2.55 (s, 2H); 3.70 (t, 2H); 4.05 (s, 2H); 5.80 (s, 1H); 7.10 (s, 2H); 7.65 (s, 1H).

PREPARATION 32

1-(N-Benzyloxycarbonyl)-4-(1,2,4-triazol-4-yl)piperidine

To a solution of 1-(N-benzyloxycarbonyl)-4-keto-piperidine (5.0 g) in methanol (25 ml) was added ammonium acetate (16.5 g) and sodium cyanoborohydride (0.94 g) and the mixture was stirred at room temperature for 24 hours. Solvent was then removed under reduced pressure and the residue partitioned between ethyl acetate and 1M sodium hydroxide solution. The ethyl acetate layer was separated, dried over magnesium sulphate, and evaporated under reduced pressure to give a yellow oil. Chromatography of this residue on silica-gel, eluting with methylene chloride-methanol-concentrated aqueous ammonia (95:5:1) gave 4-amino-1-(N-benzyloxycarbonyl)-piperidine as a yellow oil. A solution of this product (1.5 g) in toluene (20 ml) was treated with dimethylformamide azine (1.0 g) and *p*-toluenesulphonic acid (0.1 g), and the mixture was heated under reflux for 24 hours. Solvent was then removed under reduced pressure and the residue was chromatographed on silica-gel, eluting with methylene chloride-methanol-concentrated aqueous ammonia (93:7:1), to give the title compound as a colourless oil. m/e (MH^+) 287

N.M.R. (DMSO-d_6) δ = 1.82 (m, 2H); 2.01 (m, 2H); 2.83-3.09 (m, 2H); 4.12 (m, 2H); 4.40 (m, 1H); 5.11 (s, 2H); 7.28-7.45 (m, 5H); 8.65 (s, 2H).

PREPARATION 33

1-(N-t-Butoxycarbonyl-(S)-valyl)-3-(imidazol-1-yl)azetidine

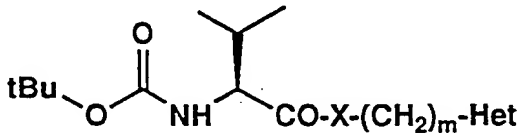
A solution of 1-(N-t-butoxycarbonyl)-3-(imidazol-1-yl)azetidine (from Preparation 18) (0.72 g) in methylene chloride (30 ml) was saturated with hydrogen chloride at 0°C and kept at this temperature for a further 1 hour. The solvent was removed under vacuum to give the amine hydrochloride which was dissolved in N,N-dimethylformamide (25 ml) and the solution was

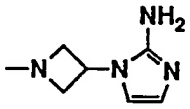
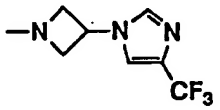
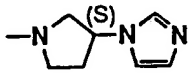
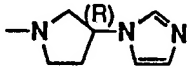
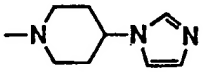
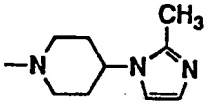
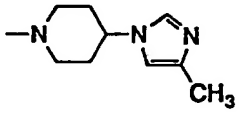
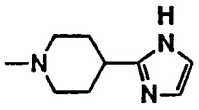
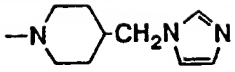
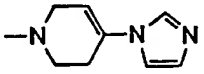
treated at room temperature with N-t-butoxycarbonyl-(S)-valine N-hydroxysuccinimide ester (1.01 g) and N,N-diisopropylethylamine (1.7 ml). The reaction mixture was stirred for 18 hours at room temperature and the solvent was then removed under vacuum. Purification of the residue by chromatography on silica gel, eluting with methylene chloride-methanol-concentrated ammonia (97:7:1) gave the title compound as a colourless foam (0.79 g). m/e 323 $(MH)^+$

N.M.R. (DMSO-d_6) δ = 0.89 (m, 6H); 1.38 (s, 9H); 1.90 (m, 1H): 3.61-3.78 (m, 1H); 3.92-4.10 (m, 1H); 4.24-4.80 (m, 3H); 5.19 (m, 1H); 6.98 (s, 1H); 7.06 (dd, 1H); 7.38 (s, 1H); 7.77 (s 1H).

PREPARATIONs 34-43

The following compounds of formula (VII) where R^4 is (S)-isopropyl and R^7 and R^8 are hydrogen were prepared following the procedure of Preparation 33 using the appropriate intermediate from Preparations 18 to 32 and coupling to N-t-butoxycarbonyl-(S)-valine N-hydroxysuccinimide ester.



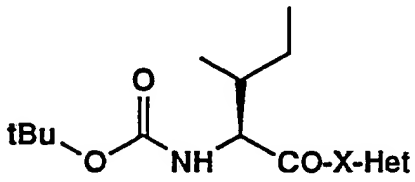
Prep No	-X-$(CH_2)_m$ Het	MH^+	Analysis % (Theoretical in brackets) C	H	N
34		338			
35		391	52.09 (52.32	6.73 6.46	14.03 14.36)
36		337	58.76 (59.11	8.51 8.46	16.63 16.22)[1]
37		337	59.46 (59.54	8.54 8.24	15.96 16.24)[2]
38		351			
39		365	61.1 (61.1	9.23 8.91	14.9 15.0)[1]
40		364.9	61.9 (61.9	9.15 8.9	14.9 15.2)[3]
41		351.1			
42		365	59.29 (59.44	8.15 8.43	14.29 14.37)[4]
43		349	61.51 (61.46	8.11 8.03	15.68 15.89)[5]

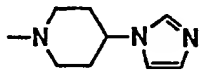
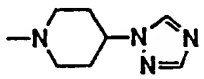
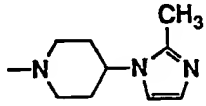
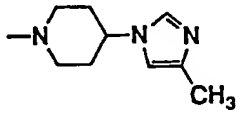
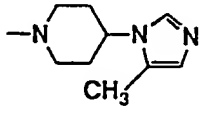
1. Hemihydrate 3. 0.25 mole H_2O 5. 0.05 mole CH_2Cl_2

2. 0.1 mole CH_2Cl_2 4. 0.3 mole CH_2Cl_2

PREPARATIONS 44-48

The following compounds of formula (VII) where R^4 is sec-butyl and m is O were prepared following the production of Preparation 33 using N-t-butoxycarbonyl-(S)-isoleucine N-hydroxy-succinimide ester in the coupling step.



Prep No	-X-Het	MH^+	Analysis % (Theoretical in brackets) C	H	N
44		365	62.01 (61.99	9.28 8.87	14.94 15.22)[1]
45		366.3			
46		379	61.7 (62.0	9.22 9.10	14.5 14.5)[2]
47		379.9	62.2 (62.0	8.98 9.10	14.33 14.5)[2]
48		379.9			

1. 0.2 mole H_2O
2. 0.5 mole H_2O

PREPARATION 49

1-(N-t-Butoxycarbonyl-(S)-isoleucyl)-4-ketopiperidine

The title compound was prepared using the same procedure as described for Preparation 33 but using 4-ketopiperidine hydrochloride hydrate instead of 3-(imidazol-1-yl)azetidine hydrochloride and (S)-isoleucine N-hydroxysuccinimide ester. m/e 313 $(MH)^+$

$[\alpha]_D^{25}$ -16° (c = 0.34%, MeOH)

N.M.R. ($CDCl_3$) δ = 0.9 (m, 6H); 1.2 (m, 1H); 1.4 (s, 9H); 1.6 (m, 1H); 1.75 (m, 1H); 2.5 (m, 4H); 3.7 (m, 2H); 4.15 (m, 2H); 4.55 (m, 1H); 5.2 (d, 2H).

PREPARATION 50

N-((R)-2-Benzyl-(S)-5-t-butoxycarbonylamino-(S)-4-(t-butyldimethylsilyloxy)-6-phenylhexanoyl)-(S)-valine

The title compound was prepared by the method described in S. J. deSolms *et al.*, J. Med Chem., 1991, 34, 2852.

PREPARATION 51

1-Isocyano-3-methyl-1-(p-toluenesulphonyl)-but-1-ene

The title compound was prepared from p-toluenesulphonylmethylisocyanide and isobutyraldehyde by the method of Van Leusen, Schaart and Van Leusen Recueil, 98, No. 5, 258 (1979). i.r. (Nujol) 2100cm^{-1}

m/e 267 $(M+ NH_3)^+$

N.M.R. ($CDCl_3$) δ = 1.15 (d, 6H); 2.5 (s, 3H); 2.84 (m, 1H); 6.88 (d, 1H); 7.4 (d, 2H); 7.85 (d, 2H).

PREPARATION 52

3-Oxetanyloxycarbonyloxysuccinimide

Oxetan-3-ol (2.0 g) and N,N-diisopropylethylamine (7.76 g) were dissolved in methylene chloride (50 ml) and the solution was added dropwise to a solution of bis-trichloromethyl carbonate (2.69 g) in methylene chloride (100 ml) maintained at -20°C over a period of

15 minutes under an atmosphere of nitrogen. The solution was then stirred for a further 15 minutes at -20°C and N-hydroxy-succinimide (3.45 g) was added in one portion. The solution was allowed to warm to room temperature over a period of 2 hours and then washed with water (50 ml), saturated aqueous sodium bicarbonate (50 ml) and brine (25 ml). The organic layer was then dried (MgSO4), filtered and the solvent was removed under vacuum to give the title compound as a light brown oil, (4.65 g). N.M.R. ($CDCl_3$) δ = 2.83 (s, 4H); 4.75 (m, 2H); 4.90 (m, 2H); 5.58 (m, 1H).

EXAMPLE 1

1-[N-((R)-2-Benzyl-(S)-5-(t-butoxycarbonylamino)-(S)-4-hydroxy- 6-phenylhexanoyl)-(S)-valyl]-3-(imidazol-1-yl)azetidine

a) 1-[N-((R)-2-Benzyl-(S)-5-(t-butoxycarbonylamino)-(S)-4-t-butyldimethylsilyloxy-6-phenylhexanoyl)-(S)-valyl]-3-(imidazol-1-yl)azetidine

A solution of 1-(N-t-butyloxycarbonyl-(S)-valyl)-3-(imidazol-1-yl)azetidine (from Preparation 33, 0.79 g) in methylene chloride (50 ml) was saturated with hydrogen chloride at 0° and kept at this temperature for a further 1 hour. The solvent was evaporated under vacuum to give the amine as a colourless solid. A solution of this product in dimethylformamide (20 ml) was added to an active ester solution previously prepared by stirring together (R)-2-benzyl-(S)-5-t-butoxycarbonylamino-(S)-4-t-butyldimethylsilyloxy)-6-phenylhexanoic acid (from Preparation 2, 1.32 g), 1-hydroxy-benzotriazole (0.36 g), 1-(3-dimethylamino-propyl)-3-ethyl-carbodiimide hydrochloride (0.58 g) and N,N-diisopropylethylamine (1.75 ml) in dimethyl-formamide (75 ml) for 20 minutes. After stirring for a further 24 hours, the solvent was removed under vacuum and the residue partitioned between ethyl acetate and water. The ethyl acetate layer was separated, dried ($MgSO_4$) and removed under vacuum to yield a colourless oil. Purification by chromatography on silica-gel eluting with ethyl acetate-methanol-concentrated aqueous ammonia (90:10:1) gave the title compound as a colourless foam (1.1 g). Found: C,66.04; H,8.26; N,9.61. $C_{41}H_{61}N_5O_5$ Si. 1/2 H_2O requires C,66.39; H,8.36; N,9.44%.

m/e 733 $(MH)^+$

$[\alpha]_D^{25}$ -6° (c = 0.29%, MeOH)

N.M.R. (DMSO-d_6) δ = 0.12(m,6H); 0.90(m,15H); 1.25-1.29(2 x s,9H); 1.96(m,1H); 2.32-2.77(m,4H);

2.89(m,1H); 3.48-3.74(m,2H); 3.88-4.06(m,3H); 4.18-4.66(m,4H); 5.12(m,1H); 6.77(t,1H); 6.98(s,1H); 7.07-7.32(m,10H); 7.40-7.82(2 x s,1H); 7.76-7.96(2 x s,1H); 8.0-8.20(2 x d,1H).

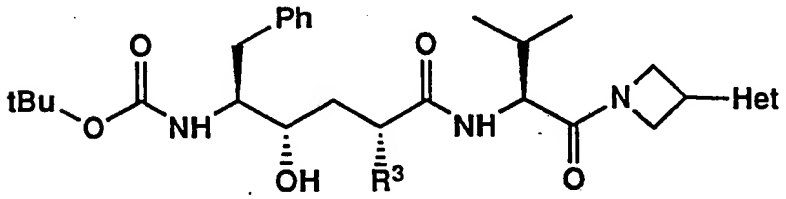
b) 1-[N-((R)-2-Benzyl-(S)-5-(t-butoxycarbonylamino)-(S)-4-hydroxy-6-phenylhexanoyl)-(S)-valyl]-3-(imidazol-1-yl)azetidine

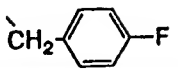
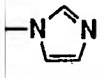
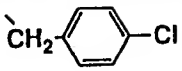
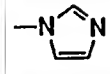
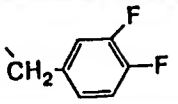
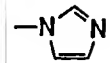
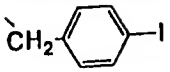
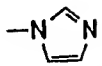
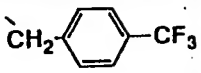
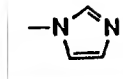
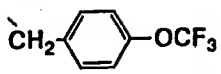
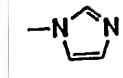
The product from step (a) (1.19 g) was dissolved in tetrahydrofuran and treated with a 1M solution of tetra-n-butylammonium fluoride in tetrahydrofuran at room temperature. After 48 hours the solvent was removed under vacuum, the product taken up in ethyl acetate, washed with saturated aqueous sodium bicarbonate and water, dried over $MgSO_4$ and the solvent evaporated under vacuum. Purification by chromatography on silica-gel eluting with methylene chloride-methanol-concentrated aqueous ammonia (93:7:1) followed by recrystallisation from ethyl acetate/hexane gave the product as a colourless solid, (0.52 g), m.p. 124-126°. Found: C,67.99; H,8.01; N,11.06. $C_{35}H_{47}N_5O_5$ requires C,68.05; H,7.67; N,11.34%. m/e 618 $(MH)^+$ $[\alpha]_D^{25}$ -3° (c = 0.14%, MeOH)

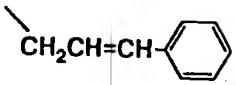
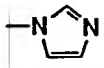
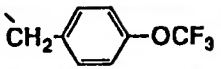
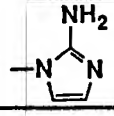
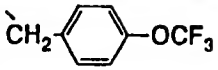
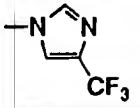
N.M.R. (DMSO-d_6) δ = 0.80(m,6H); 1.21-1.47(m,1H); 1.27(s,9H); 1.58-1.71(m,1H); 1.81-1.98(m,1H); 2.44-2.62(m,2H); 2.68-2.92(m,3H); 3.37-3.61(m,2H); 3.85-4.02(m,2H); 4.13-4.36(m,2H); 4.41-4.56(m,2H); 5.18(m,1H); 6.43(d,1H); 6.98(s,1H); 7.05-7.30(m,10H); 7.26-7.77(2 x s,1H); 7.38-7.82(2 x s,1H); 7.88-7.98(2 x d,1H).

EXAMPLES 2-10

The following compounds were prepared following the general procedure described in Example 1 but using the appropriate protected carboxylic acid intermediate of formula (IV) and the appropriate amine of formula (VIII) in the coupling step (a) followed by removal of the t-butyldimethylsilyl protecting group as described in step (b).



Ex No	R^3	Het	m.p.°C	$[\alpha]_D^{25}$	Analysis % (Theoretical in brackets) C	H	N
2			184-186°	+2°	66.3 (66.1	7.38 7.29	11.0 11.0
3			159-160°	+7°	64.5 (64.5	7.24 7.11	10.58 10.7
4			163-165°		64.5 (64.3	7.01 6.94	10.4 10.7)
5			210-212°	+21°	57.0 (56.5	6.36 6.23	9.41 9.41)
6			209-211°	+1°	63.4 (63.1	6.88 6.76	10.2 10.2)
7			210-213°	-2°	62.0 (61.7	6.80 6.60	10.0 10.0)

Ex No	R^3	Het	m.p.°C	$[\alpha]_D^{25}$	Analysis % (Theoretical inbrackets) C	H	N
8			170-172°	+53°	69.03 (69.03	7.64 7.67	10.95 10.88)
9			-	+6°	59.4 (59.6	6.63 6.67	11.5 11.6)
10			148-150°	-	57.98 (57.73	5.96 5.89	8.90 9.09)

EXAMPLE 11

1-[N-((S)-5-(t-Butoxycarbonylamino)-(S)-4-hydroxy-6-phenyl-(R)- 2-(4-(trifluoromethoxy)benzyl)hexanoyl)-(S)-valyl]-(S)-3-(imidazol-1-yl)pyrrolidine

The procedure of Example 1 was followed using 1-(N-t-butoxycarbonyl-(S)-valyl)-3(S)-imidazol-1-yl)pyrrolidine (Preparation 36) in the coupling step followed by removal of the t-butyldimethylsilyl group as described in Example 1(b) to give the title product. m.p. 111-112°C. Found: C, 61.90; H, 6.85; N, 9.61. $C_{37}H_{48}F_3N_5O_6$ requires C, 62.08; H, 6.76; N, 9.78%. m/e $(MH)^+$ 716

$[\alpha]_D^{25}$ + 2° (c = 0.1%, MeOH) $[\alpha]_{365}^{25}$ + 14° (c = 0.1%, MeOH)

N.M.R. (DMSO-d6) δ = 0.65-0.90 (m, 6H); 1.15-1.35 (m, 10H); 1.60 (m, 1H); 1.85 (m, 1H); 2.05-2.90 (m, 7H); 3.20-4.10 (m, 6H); 4.20 (m, 1H); 4.55 (m, 1H); 4.85 (m, 1H); 6.40 (d, 1H); 6.90-6.95 (2 x s, 1H); 7.10-7.30 (m, 10H); 7.70-7.80 (2 x s, 1H); 7.90-8.0 (2 x d, 1H).

EXAMPLE 12

1-[N-((S)-5-(t-Butoxycarbonylamino)-(S)-4-hydroxy-6-phenyl-(R)- 2-(4-(trifluoromethoxy)benzyl)hexanoyl)-(S)-valyl]-(R)-3-(imidazol-1-yl)pyrrolidine

The above procedure was followed but starting with 1-(N-t-butoxycarbonyl-(S)-valyl-3(R)-imidazol-1-yl)pyrroldine to give the title product, m.p. 109°C. Found: C, 61.44; H, 7.05; N, 9.71. $C_{37}H_{48}F_3N_5O_6$ $^2/_5$ H_2O requires C, 61.47; H, 6.80; N, 9.69%. m/e $(MH)^+$ 716

$[\alpha]_D^{25}$ -23° (c = 0.1%, MeOH)

N.M.R. (DMSO-d6) δ = 0.70-0.85 (m, 6H); 1.15-1.40 (m, 10H); 1.60 (m, 1H); 1.85 (m, 1H); 2.00-2.90 (m, 7H); 3.25-3.70 (m, 5H); 3.95 (m, 1H); 4.20 (m, 1H); 4.45-4.65 (m, 1H); 4.75-4.95 (2 x m, 1H); 6.40 (m, 1H); 6.90 (s, 1H); 7.05-7.25 (m, 10H); 7.70-7.75 (2 x s, 1H); 7.90-8.05 (2 x d, 1H).

EXAMPLE 13

1-[N-((R)-2-Benzyl-(S)-5-(t-butoxycarbonylamino)-(S)-4-hydroxy-6-phenylhexanoyl)-(S)-valyl]-4-(imidazol-1-yl)piperidine

a) 1-[N-((R)-2-Benzyl-(S)-5-(t-butoxycarbonylamino-(S)-4-t-butyldimethylsilyloxy-6-phenylhexanoyl)-(S)-valyl-4-(imidazol-1-yl)piperidine

The title compound was prepared from 1-(N-t-butoxycarbonyl-(S)-valyl-4-imidazol-1-ylpiperidine and (S)-5-t-butoxycarbonylamino-(S)-4-t-butyldimethyl-silyloxy-(R)-2-benzyl-6-phenylhexanoic acid using the same procedure as described in Example 1 step (a). Purification by chromatography on silica-gel eluting with ethyl acetate-methanol-concentrated aqueous ammonia (90:10:1) gave the product as a colourless foam (0.71 g). Found: C,67.32; H,8.44; N,9.22. $C_{43}H_{65}N_5O_5$ Si. 1/4 H_2O requires C,67.54; H,8.56; N,9.15%.

m/e 760 $(MH)^+$

N.M.R. (DMSO-d_6) δ = 0.12(m,6H); 0.91(m,15H); 1.13-1.30(m,2H); 1.26(s,9H); 1.47-1.76(m,2H); 1.89-2.08(m,4H); 2.36-2.97(m,6H); 3.50-3.75(m,2H); 3.99-4.61(m,4H); 6.79(m,1H); 6.90(d,1H); 7.07-7.29(m,11H); 7.68-7.73(2 x s,1H); 7.93-7.99(2 x d,1H).

b) 1-[N-(R)-2-Benzyl-(S)-5-(t-butoxycarbonylamino)-(S)-4-hydroxy-6-phenylhexanoyl)-(S)-valyl]-4-(imidazol-1-yl)piperidine

The title compound was prepared from the product of step a) above by the same procedure as described for Example 1 step b). Purification by chromatography on silica-gel eluting with methylene chloride-methanol-concentrated aqueous ammonia followed by recrystallisation from ethyl acetate/hexane gave the product as a colourless solid, m.p. 121-123° (0.48 g). Found: C,68.38; H,7.96; N,10.59. $C_{37}H_{51}N_5O_5$. 1/4 H_2O requires C,68.30; H,7.92; N,10.77%.

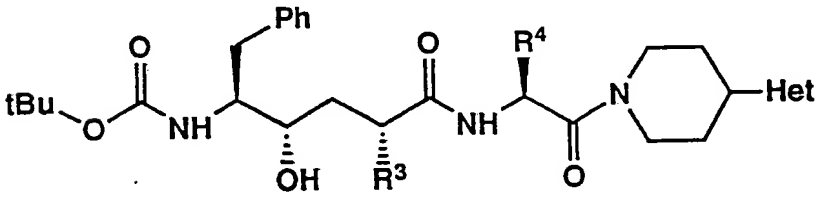
m/e 646 $(MH)^+$

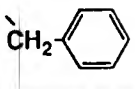
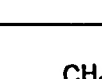
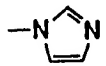
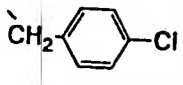
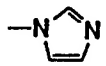
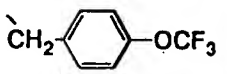
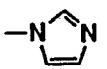
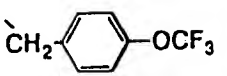
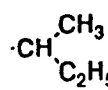
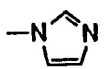
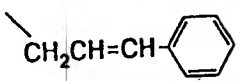
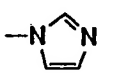
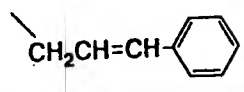
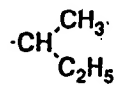
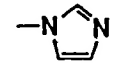
$[\alpha]^{25}$ -10° (c = 0.1, MeOH)

N.M.R. (DMSO-d_6) δ = 0.77(m,6H); 1.19-1.37(m,1H); 1.28(s,9H); 1.48-1.72(m,3H); 1.82-2.04(m,3H); 2.42-2.94(m,7H); 3.38-3.60(m,2H)); 3.96-4.15(m,1H); 4.28(m,1H); 4.37-4.58(m,3H); 6.43(d,1H); 6.88(s,1H); 7.04-7.29(m,11H); 7.65-7.69(2 x s,1H); 7.84-7.88(2 x d,1H).

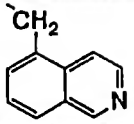
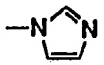
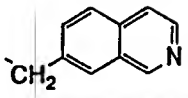
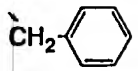
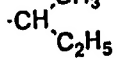
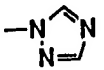
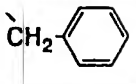
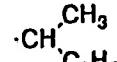
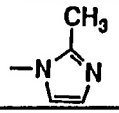
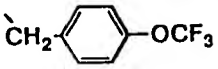
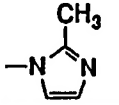
Examples 14-37

The following compounds were prepared following the procedure described in Example 13 but using the appropriate protected carboxylic acid intermediate of formula (IV) and the appropriate valine or isoleucine derivative of formula (VIII) in the coupling step (a) followed by removal of the t-butyldimethylsilyl protecting group as described in step (b).

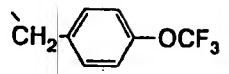
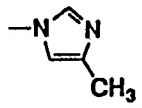
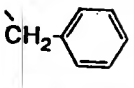
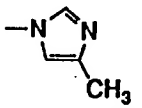
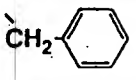
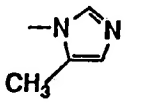
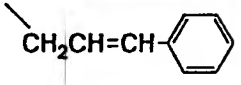
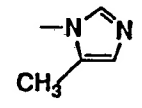
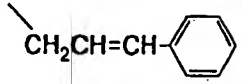
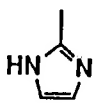
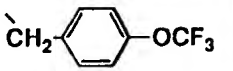
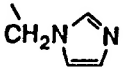


Ex No	R^3	R^4	Het	m.p °C	[α]	Analysis % C (Theoretical in brackets)	H	N
14				-	-10 (c = 0.13%)	65.35 (69.17	8.35 8.10	10.53 10.61)
15		—$CH(CH_3)_2$		113-115	+11	64.4 (64.3	7.76 7.59	10.00 10.14)
16		—$CH(CH_3)_2$		104-107	-4	62.01 (62.08	7.09 7.03	9.19 9.05)
17				150-152	+6	62.30 (62.21	6.97 6.97	9.03 9.03)[1]
18		—$CH(CH_3)_2$		125-126	+20	68.78 (69.72	7.94 7.50	10.31 10.42)
19				143-144	+28	70.05 (70.04	8.36 8.08	10.07 10.21)

1 hemihydrate

Ex No	R^3	R^4	Het	m.p °C	[α]	Analysis % C (Theoretical in brackets)	H	N
20		$-CH(CH_3)_2$		122-125	+5	68.19 (68.06	7.76 7.57	11.58 11.91)
21		$-CH(CH_3)_2$		116-119	+17	64.4 (64.3	7.76 7.59	10.00 10.14)
22				-	-6°	67.6 (67.3	7.84 7.93	12.7 12.7)
23				-	-2.8°	70.0 (70.3	8.69 7.82	9.75 9.76)[2]
24		$-CH(CH_3)_2$			0°	63.2 (63.0	7.28 7.05	9.12 9.41)

2. 0.5 mole C_6H_{14}

Ex No	R^3	R^4	Het	m.p °C	[α]	Analysis % C (Theoretical in brackets)	H	N
25		$-CH(CH_3)_2$			+6°	62.8 (63.0	7.02 7.05	9.35 9.41)
26					-6°	69.6 (69.5	8.56 8.23	10.3 10.4)
27					-11°	69.4 (69.5	8.56 8.23	10.0 10.3)
28					-11°	69.4 (69.5	8.56 8.23	10.0 10.3)
29		$-CH(CH_3)_2$		199-201	-	69.7 (69.7	8.08 7.95	10.3 10.4)
30		$-CH(CH_3)_2$		169	+2°	57.00 (57.20	6.71 6.59	7.67 7.76)[3]

3. Tartrate salt, hemihydrate

EXAMPLE 31

1-[N-((R)-2-Benzyl-(S)-5-(t-butoxycarbonylamino)-(S)-4-hydroxy-6-phenylhexanoyl)-(S)-valyl]-4-(1,2,4-triazol-4-yl)piperidine

The benzyloxycarbonyl protecting group was removed from 1-(N-benzyloxycarbonyl)-4-(1,2,4-triazol-4-yl)-piperidine (Preparation 32) by catalytic hydrogenation and the resulting amine product coupled to N-((R)-2-benzyl-(S)-5-t-butoxycarbonylamino-(S)-4-t-butyldimethylsilyloxy)-6-phenylhexanoyl)-(S)-valine following the procedure described for Example 1 step (a), to give 1-[N-((R)-2-benzyl-(S)-5-(t-butoxy-carbonylamino)-(S)-4-(t-butyldimethylsilyloxy)-6-phenylhexanoyl)-(S)-valyl]-4-(1,2,4-triazol-4-yl)piperidine, m/e 761 (MH^+).

The above product was treated with tetra-n-butylammonium fluoride following the procedure of Example 1 step (b) to give the title product. Found: C,64.90; H,7.93; N,12.48. $C_{36}H_{50}N_6O_5 . H_2O$ requires C,65.06; H,7.83; N,12.65%. m/e 647 $(MH)^+$

$[\alpha]_D^{25}$ -9° (c = 0.12 MeOH)

N.M.R. (DMSO-d_6) δ = 0.70-0.92(m,6H); 1.15-1.41(m,10H); 1.46-1.76(m,3H); 1.81-2.16(m,3H); 2.53-2.96(m,7H); 3.38-3.64(m,2H); 3.95-4.15(m,1H); 4.27-4.61(m,4H); 6.36-6.44(m,1H); 7.01-7.12(m,10H); 7.78-7.91(m,1H); 8.54(s,1H); 8.60(s,1H).

EXAMPLE 32

1-[N-((S)-5-(t-Butoxycarbonylamino)-(S)-4-hydroxy-6-phenyl-(R)- 2-(4-trifluoromethoxybenzyl)hexanoyl)-(S)-valyl]-4-(imidazol-1-yl)1,2,5,6-tetrahydropyridine; tartrate

a) Reaction of (S)-5-t-butoxycarbonylamino-(S)-4-t butyldimethylsilyloxy)-(R)-2-(4-trifluoromethoxy-benzyl)-6 phenylhexanoic acid and the amine prepared by deprotection of 1-(N-t-butoxycarbonyl)-4-imidazol-2-

yl(1,2,5,6-tetrahydropyridine) following the procedure of Example 1(a) gave 1-[N-((S)-5-t-butyloxycarbonyl-amino)-(S)-4-t-butyldimethylsilyloxy)-6-phenyl-(R)-2-(4-trifluoromethoxybenzyl)hexanoyl-(S)-valyl]-4-(imidazol-1-yl)-1,2,5,6-tetrahydropyridine. Found: C,62.19; H,7.39; N,8.38. $C_{33}H_{62}F_3N_5O_6Si$ 1/10 CH_2Cl_2 requires C,62.27; H,7.37; N,8.23%.

m/e 842 $(M)^+$

$[\alpha]_D^{25}$ -10° (c = 0.1%, MeOH)

N.M.R. ($CDCl_3$) δ = 0.10 (m,6H); 0.80 (d,6H); 0.90(s,9H); 1.35(s,9H); 1.20-1.95(m,5H); 2.35-2.55(m,3H); 2.70(m,2H); 3.41-4.10(m,6H); 4.55(m,1H); 4.65(d,1H); 5.60-5.80(2 x s, 1H); 6.25(d,1H); 6.90-7.30(m,11H); 7.65(s,1H).

b) Deprotection by reaction with tetra-n-butylammonium fluoride following the procedure of Example 1(b) gave the title product, free base. This was dissolved in absolute ethanol and treated with a solution of l-tartaric acid (0.14 g) in absolute ethanol. Addition of ether gave a precipitate which was filtered and dried to give the l-tartrate salt as a colourless solid, m.p. 92-152°C (0.61 g). Found: C, 57.40; H, 6.64; N, 7.51. $C_{38}H_{38}F_3N_5O_6.CH_{16}O_6$ $^1/_3$ H_2O requires C, 57.11; H, 6.23; N, 7.93%.

m/e $(MH)^+$ 728

$[\alpha]_D^{25}$ +6.7° (c = 0.1%, MeOH)

$[\alpha]_{365}^{25}$ - 6.7° (c = 0.1%, MeOH)

N.M.R. (DMSO-d6) δ = 0.75 (m, 6H); 1.25 (s, 9H); 0.95-1.40 (m, 1H); 1.50 (m, 1H); 1.90 (m, 1H); 2.35-2.95 (m, 7H); 3.0-3.85 (m, 4H); 4.0 (m, 1H); 4.10 (m, 1H); 4.25 (s, 2H); 4.40-4.60 (m, 2H); 6.0 (m, 1H); 6.40 (d, 1H); 7.0 (s, 1H); 7.10-7.30 (m, 9H); 7.50 (s, 1H); 7.95 (m, 2H).

EXAMPLE 33

1-[N-((S)-5-(t-Butoxycarbonylamino)-(S)-4-hydroxy-6-phenyl-(R)-2-(4-trifluoromethoxybenzyl)hexanoyl)-(S)-isoleucyl]-4-(5-isopropylimidazol-1-yl)piperidine

a) Deprotection of 1-(N-t-butoxycarbonyl-(S)-isoleucyl)-4-ketopiperidine (Preparation 49) followed by reaction with (S)-5-t-butoxycarbonylamino-(S)-4-(t-butyldimethylsilyloxy)-6-phenyl-(R)-2-(4-trifluoromethoxybenzyl)hexanoic acid (Preparation 10) gave 1-[N-((S)-5-(t-butoxycarbonylamino-(S)-4-(t-butyldimethylsilyloxy)-6-phenyl-(R)-2-(4-trifluoromethoxybenzyl)hexanoyl-(S)-isoleucyl]-4-ketopiperidine. Found: C,62.50; H,8.00; N,5.20. $C_{42}H_{62}F_{33}O_7Si$ requires C,62.60; H,7.80; N,5.20%. m/e 806.5 $(MH)^+$. $[\alpha]_D^{25}$ -5.30 (c = 0.1%, MeOH).

b) The above product (2.0 g) and aqueous ammonia (specific gravity 0.88), in ethanol (25 ml) were hydrogenated with palladium on carbon (5 100 mg) at 30 p.s.i. (2.0 bar) for 4 hours. Filtration of catalyst and evaporation of solvent gave a colourless foam, 2.0 g, which was chromatographed on silica-gel, eluting with dichloromethane:methanol:concentrated aqueous ammonia (90:10:1), to give, after concentration of the appropriate fractions 1-[N-((S)-5-t-butoxycarbonyl-amino-(S)-4-t-butyldimethylsilyloxy-6-phenyl-(R)-2-(4-trifluoromethoxy)benzyl)hexanoyl)-(S)-isoleucyl]-4-aminopiperidine as a colourless glass, 1.53 g.

c) The product from step b) above (404 mg) and 1-isocyano-3-methyl-1-(p-toluenesulphonyl)-but-1-ene, (from Preparation 51, 150 mg) were stirred together in methanol (15 ml) with diisopropylethylamine (100 mg) for 16 hours. The solvent was removed at 40°C and the residue purified by chromatography on silica-gel, eluting with dichloromethane:methanol:concentrated aqueous ammonia (98:2:0.4), to give 1-[N-((S)-5-(t-

butoxycarbonylamino)-(S)-4-t-butyldimethylsilyoxy-6-phenyl-(R)-2-(4-trifluoromethoxybenzyl)hexanoyl)-(S)-isoleucyl]-4-(5-isopropylimidazolyl-1-yl)piperidine as a colourless glass (380 mg). m/e 900 (MH^+).

d) The product from step c) above was deprotected by treatment with tetra-n-butylammonium fluoride following the procedure of Example (1b) to give the title product.

Found : C, 63.5; H, 7.70; N, 8.70. $C_{42}H_{58}F_3N_5O_6$. $^1/_2H_2O$ requires. C, 63.5; H, 7.48; N, 8.81%.

m/e 785.9 $(MH)^+$

N.M.R. (DMSO-d_6) δ = 0.8 (m, 6H); 1.05 (m, 1H); 1.2 (m, 6H); 1.3 (s, 9H); 1.3-2.0 (m, 7H); 2.5-3.0 (m, 6.5H); 3.2 (bt, 0.5H); 3.3 (m, 1H); 3.58 (m, 1H); 4.13 (m, 3H); 4.4-4.65 (m, 4H); 6.43 (m, 1H); 6.63 (s, 1H); 7.1-7.3 (m, 9H); 7.47, 7.6 (s,s, 1H); 7.95 (bt, 1H).

EXAMPLE 34

1-[N-((S)-4-Hydroxy-(S)-5-(oxetan-3-yloxycarbonyl-amino)-6-phenyl-(R)-2-(4-trifluoromethoxybenzyl)-hexanoyl)-(S)-valyl]-4-(imidazol-1-yl)piperidine

The product from Example 16 (4.0 g) was dissolved in methylene chloride (40 ml) and cooled in an ice bath. Trifluoroacetic acid (10 ml) was added dropwise over a period of 5 minutes and the solution stirred at 0°C for 1.5 hours. The solvent was evaporated under vacuum and the residue taken up in ethyl acetate (250 ml) and washed with 1M sodium hydroxide (50 ml) and brine (50 ml). The organic solution was dried ($MgSO_4$), filtered and the solvent was evaporated under vacuum. Chromatography on silica gel, eluting with ethyl acetate-methanol-concentrated aqueous ammonia (90:10:1) gave 1-[N-(S)-5-amino-(S)-4-hydroxy-6-phenyl-(R)-2-(4-trifluoromethoxybenzyl)hexanoyl)-(S)-valyl]-4-(imidazol-1-yl)piperidine as a white solid (2.98 g).

m/e 630 $(MH)^+$

N.M.R. (DMSO-d_6) δ = 0.78(m,6H); 1.34-1.75(m,4H); 1.81-2.07(m,3H); 2.37-2.93(m,8H); 3.05-3.29(m,3H); 4.05-4.56(m,5H); 6.88(s,1H); 7.10-7.27(m,10H); 7.67(d,1H); 7.95(m,1H);

b) The product from step a) (0.818 g) was dissolved in methylene chloride (30 ml). 3-Oxetanylcarbonyloxy-succinimide (0.344 g) was added and the solution stirred at room temperature for one hour. The solution was then washed with 0.5M sodium hydroxide (15 ml) and brine (15 ml), dried ($MgSO_4$), filtered and evaporated under vacuum. The resulting solid was recrystallised from ethyl acetate to give the title compound as a white solid, m.p. 201-203°C. Found: C,60.32; H,6.22; N,9.15. $C_{37}H_{46}F_3N_5O_7$ 1/2 H_2O requires C,60.15; H,6.41; N,9.48%.

m/e 730 $(MH)^+$

$[\alpha]_D^{25}$ +3° (c = 0.1%, MeOH), $[\alpha]_{365}^{25}$ +32° (c = 0.1, MeOH)

N.M.R. (DMSO-d_6) δ = 0.77(m,6H); 1.22-2.05(m,7H); 2.50-2.93(m,8H); 3.42-3.60(m,2H); 4.08(d,1H); 4.22-4.73(m,7H); 5.12(m,1H); 6.89(s,1H); 7.11-7.28(m,11H); 7.67(d,1H); 7.86(d,1H).

EXAMPLE 35

1-[N-(S)-4-Hydroxy-(S)-5-(oxetan-3-yloxycarbonylamino)-6-phenyl -(R)-2-(3-phenylprop-2-enyl)hexanoyl)-(S)-isoleucyl]-4-(imidazol-1-yl)piperidine

A solution of the product from Example 18 (0.45 g) in methylene chloride (20 ml) was treated with anhydrous trifluoroacetic acid (4 ml) at 0-5°C for 5 hours. The reaction was then concentrated under vacuum and the residual gum azeotroped with toluene (x3) and dried. A solution of the crude amine product and diisopropyl-ethylamine (0.82 ml) in methylene chloride (25 ml) was then cooled to 5°C and a solution of 3-oxetanyl-oxycarbonyloxysuccinimide (0.22 g) in methylene chloride (5 ml) added dropwise. The reaction was

allowed to warm to room temperature, maintained for 17 hours and then washed with water, dried ($MgSO_4$), and evaporated under vacuum. Purification by chromatography on silica gel eluting with methylene chloride-methanol-concentrated aqueous ammonia (95:4:1) gave product as a colourless foam, m.p. 157-160°C (0.26 g).

Found : C, 66.45; H, 7.49; N, 10.22. $C_{38}H_{49}N_5O_6$ $^3/_4H_2O$ requires C, 66.60; H, 7.43; N, 10.22%.

$[\alpha]_D^{25}$ +32° (c = 0.1%, MeOH)

N.M.R. (DMSO-d_6) δ = 0.75 (m, 6H); 1.25-2.95 (m, 12H); 3.05-3.65 (m, 4H); 3.95-4.75 (m, 10H); 5.05 (m, 1H); 6.10 (m, 1H); 6.30 (d, 1H); 6.80-6.85 (2 x s, 1H); 6.95-7.35 (m, 11H); 7.55-7.65 (2 x s, 1H); 7.95 (d, 1H).

EXAMPLE 36

1-[N-(S)-4-Hydroxy-(S)-5-(isopropyloxycarbonylamino)-6-phenyl-(R)-2-(3-phenylprop-2-enyl)hexanoyl)-(S)-isoleucyl]-4-(imidazol-1-yl)piperidine; tartrate

The title compound was prepared by the procedure described above for Example 35, except that isopropylchloroformate was used for reaction with the amine intermediate. Purification by chromatography on silica gel eluting with methylene chloride-methanol-concentrated aqueous ammonia (97:2:1 to 95:4:1) gave the product as a colourless powder, which was recrystallised from ethyl acetate-hexane. The free base was dissolved in ethanol and treated with a solution of l-tartaric acid. Addition of diethyl ether gave the tartrate salt as a colourless powder, m.p. 156-157°C.

Found : C, 61.60; H, 6.87; N, 8.38. $C_{38}H_{51}N_5O_5$ $C_4H_6O_6$ $^1/_2$ H_2O requires C, 61.75; H, 7.16; N, 8.57%.

m/e 658 $(MH)^+$

$[\alpha]_D^{25}$ +26° (c = 0.1%, MeOH)

N.M.R. (DMSO-d_6) δ = 0.75 (m, 6H); 1.0 (m, 6H); 1.25-3.65 (m, 14H); 3.95-4.70 (m, 10H); 6.10 (m, 1H); 6.30 (d, 1H); 6.65 (d, 1H); 6.85-6.90 (2 x s, 1H); 7.05-7. (m, 11H); 7.65-7.75 (2 x s, 1H); 7.90 (d, 1H).

EXAMPLE 37

1-[N-(S)-5-(t-Butoxycarbonylamino)-6-phenyl-(R)-2-(4-trifluoromethoxybenzyl)-4-((S)-valyloxy)hexanoyl)-(S)-valyl]-4-(imidazol-1-yl)piperidine; tartrate

a) N-Benzyloxycarbonyl-L-valine (1.03 g) and dicyclohexylcarbodiimide (0.51 g) were dissolved in dichloromethane (25 ml) and the mixture was stirred for 3 hours. The precipitated dicyclohexylurea was removed by filtration and the filtrate evaporated to a white foam. This white foam was combined with the product of Example 16 in N,N-dimethylformamide (20 ml) and 4-dimethylaminopyridine (0.025 g) added. After stirring at room temperature for five days the mixture was partitioned between ethyl acetate and water. Drying of the organic extract ($MgSO_4$) and evaporation of the solvent followed by silica-gel chromatography, eluting with ethyl acetate:methanol (0-10%) gave 1-[N-(S)-4-(N-benzyloxycarbonyl-(S)-valyloxy)-(S)-5-(t-butoxy-carbonylamino)-6-phenyl-(R)-2-(4-(trifluoromethoxy)-benzyl)hexanoyl-(S)-valyl]-4-(imidazol-1-yl)piperidine as a white foam. Found: C,62.63; H,6.73; N,8.37. $C_{51}H_{65}F_3N_6O_9$. H_2O requires C,62.43; H,6.88; N,8.56%.
m/e 963 $(MH)^+$

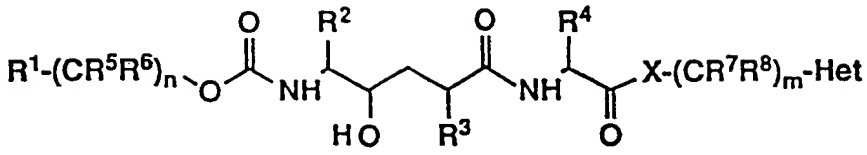
b) The product from step a) above (0.95 g) was dissolved in absolute ethanol (50 ml), and the solution was treated with 10% palladium on charcoal (0.1 g) and hydrogenated at 60p.s.i., (4.1 bar) at room temperature for 4 hours. After removal of the catalyst the filtrate was evaporated to dryness. Purification by silica-gel chromatography eluting with dichloromethane-methanol-concentrated aqueous ammonia (97:3:0.5) gave the product as a white foam. This foam was dissolved in

ethyl acetate (6 ml) and a solution of tartaric acid (0.089 g) in 10% methanol ethyl acetate (10 ml) was added. Evaporation of the solvent and trituration with diethyl ether gave the product as a white glass (0.48 g), m.p. 122°C. Found C, 56.74; H, 6.78; N, 8.56. $C_{43}H_{59}F_3N_6O_7:C_4H_6O_6:H_2O$ requires C, 56.61; H, 6.77; N, 8.43%.

N.M.R. (DMSO-d_6) δ = 0.8-0.95 (m, 12H); 1.2 (s, 11H); 1.35-1.77 (m, 2H); 1.8-2.15 (m, 4H); 2.4-3.2 (m, 8H); 3.51 (m, 1H); 3.8 (m, 1H); 4.1 (m, 1H); 4.3 (m, 1H); 4.4-4.65 (m, 2H); 4.81 (m, 1H); 6.83 (s, 1H); 7.0-7.32 (m, 12H); 7.66 (d, 1H); 7.73-8.0 (m, 1H).

CLAIMS

1. A compound having the formula:



(I)

or a pharmaceutically acceptable salt thereof or bioprecursor therefor, wherein-:

R^1 is C_1-C_6 alkyl, C_3-C_8 cycloalkyl, aryl, heterocyclyl or $CONR^9R^{10}$;

R^2 is C_1-C_6 alkyl, C_3-C_8 cycloalkyl(C_1-C_4)alkyl, aryl(C_1-C_4)alkyl or heterocyclyl(C_1-C_4)alkyl;

R^3 is C_1-C_6 alkyl, C_3-C_8 cycloalkyl, C_3-C_8 cycloalkyl(C_1-C_4)alkyl, aryl(C_1-C_4)alkyl, aryl(C_2-C_4)-alkenyl, heterocyclyl(C_1-C_4)alkyl or heterocyclyl(C_2-C_4)-alkenyl;

R^4 is C_1-C_6 alkyl, C_3-C_8 cycloalkyl, aryl or heterocyclyl;

each of R^5, R^6, R^7 and R^8 is independently H, C_1-C_6 alkyl or C_3-C_8 cycloalkyl; or R^5 and R^6, or R^7 and R^8 may be joined together to form a 3 to 8 membered carbocyclic ring;

X is a 4-10 membered mono or bicyclic heterocyclic

group containing carbon ring atoms and one ring nitrogen atom through which the group is attached to the adjacent carbonyl group; the group may be saturated or partially unsaturated and, in addition to the -$(CR^7R^8)_m$-Het substituent, it may be substituted by up to 4 further substituents each independently chosen from F, C_1-C_6 alkyl, C_3-C_8 cycloalkyl, OR^{11} or NR^9R^{10};

Het is an imidazolyl or triazolyl group either of which may optionally be substituted by C_1-C_6 alkyl, C_3-C_8 cycloalkyl, NR^9R^{10} or $CONR^9R^{10}$,

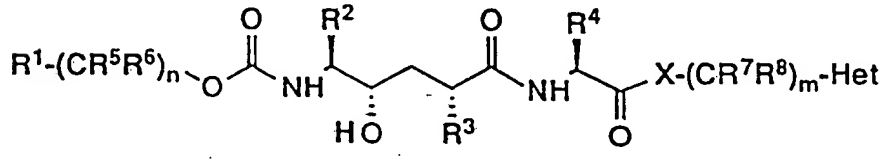
each of R^9 and R^{10} is independently H, C_1-C_6 alkyl or C_3-C_8 cycloalkyl,or R^9 and R^{10} may be joined together to form, with the nitrogen to which they are attached, a 4 to 8 membered nitrogen- containing heterocyclic group,

R^{11} is H, C_1-C_6 alkyl or C_3-C_8 cycloalkyl;

n and m are each independently 0, 1 or 2;

wherein any alkyl or cycloalkyl group included in the aforementioned definitions may optionally be fully or partially substituted by fluorine.

2. A compound as claimed in claim 1 having the stereochemistry:



wherein R^1 to R^8, n, m, X and Het are as previously defined in claim 1.

3. A compound as claimed in claim 1 or claim 2 wherein R^1 is t-butyl, isopropyl, or oxetanyl and n is 0.

4. A compound as claimed in claim 3 wherein R^2 is benzyl.

5. A compound as claimed in claim 3 or claim 4 wherein R^4 is isopropyl or sec-butyl.

6. A compound is claimed in any one of claims 3 to 5 wherein R^3 is benzyl optionally substituted in the phenyl ring by methyl, fluoro, chloro, iodo, CF_3, or OCF_3, or R^3 is 3-phenylpropyl or 3-phenyl-prop-2-enyl.

7. A compound as claimed in any one of claims 3 to 6 wherein each of R^5, R^6, R^7 and R^8 is H.

8. A compound as claimed in any one of claims 3 to 7 wherein m is 0 or 1.

9. A compound as claimed in claim 1 wherein said compound is:

1-[N-((R)-2-benzyl-(S)-5-(t-butoxycarbonylamino)-(S)-4-hydroxy-6-phenylhexanoyl)-(S)-valyl]-3-(imidazol-1-yl)azetidine,

1-[N-((R)-2-benzyl-(S)-5-(t-butoxycarbonylamino)-(S)-4-hydroxy-6-phenylhexanoyl)-(S)-valyl]-4-(imidazol-1-yl)piperidine,

1-[N-((S)-5-(t-butoxycarbonylamino)-(S)-4-hydroxy-6-phenyl-(R)-2-(3-phenylprop-2-en-1-yl)hexanoyl)-(S)-valyl]-4-(imidazol-1-yl)piperidine,

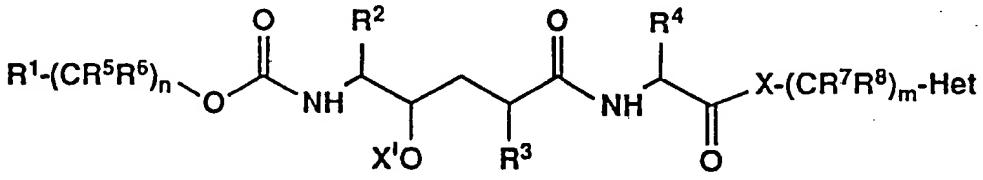
1-[N-((S)-5-(t-butoxycarbonylamino)-(S)-4-hydroxy-6-phenyl-(R)-2-(4-trifluoromethoxybenzyl)hexanoyl)-(S)-valyl]-4-(imidazol-1-yl)piperidine,

1-[N-((S)-5-(t-butoxycarbonylamino)-(R)-2-(4-chlorobenzyl)-(S)-4-hydroxy-6-phenylhexanoyl)-(S)-valyl]-3-(imidazol-1-yl)azetidine and

1-[N-((S)-5-(t-butoxycarbonylamino)-(S)-4-hydroxy-6-phenyl-(R)-2-(4-trifluoromethoxybenzyl)hexanoyl)-(S)-

isoleucyl]-4-(imidazol-1-yl)piperidine

10. A process for preparing a compound of the formula (I) as claimed in claim 1 which comprises removing the protecting groups from a compound of the formula:



(IX)

wherein X^1 is a selectively removable hydroxy-protecting group and R^1 to R^8, X and Het are as defined in claim 1, and isolating the compound of formula (I) and optionally forming a pharmaceutically acceptable salt thereof.

11. A pharmaceutical composition comprising a compound of the formula (I) or (Ia) or a pharmaceutically acceptable salt thereof or bioprecursor therefor as claimed in any one of claims 1 to 9, together with a pharmaceutically acceptable diluent or carrier.

12. A compound of the formula (I), or (Ia) or a pharmaceutically acceptable salt thereof or bioprecursor therefor, as claimed in any of claims 1 to 9 for use in medicine, in particular for use in the treatment or prophylaxis of human retroviral infections.

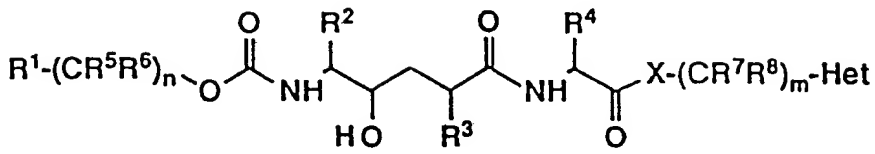
13. The use of a compound of the formula (I) or a (Ia) as claimed in any one of claims 1 to 9, or a pharmaceutically acceptable salt thereof, for the manufacture of a medicament for use in the treatment or

phrophylaxis of human retroviral infections.

14. A method of treating human retroviral infections which comprises administering an effective amount of a compound of the formula (I) or (Ia) as claimed in any one of claims 1 to 9.

PROCESS CLAIMS

15. A process for producing a compound having the formula:



(I)

or a pharmaceutically acceptable salt thereof, wherein-:

R^1 is C_1-C_6 alkyl, C_3-C_8 cycloalkyl, aryl, heterocyclyl or $CONR^9R^{10}$;

R^2 is C_1-C_6 alkyl, C_3-C_8 cycloalkyl(C_1-C_4)alkyl, aryl(C_1-C_4)alkyl or heterocyclyl(C_1-C_4)alkyl;

R^3 is C_1-C_6 alkyl, C_3-C_8 cycloalkyl, C_3-C_8 cycloalkyl(C_1-C_4)alkyl, aryl(C_1-C_4)alkyl, aryl(C_2-C_4)-alkenyl, heterocyclyl(C_1-C_4)alkyl or heterocyclyl(C_2-C_4)-alkenyl;

R^4 is C_1-C_6 alkyl, C_3-C_8 cycloalkyl, aryl or heterocyclyl;

each of R^5, R^6, R^7 and R^8 is independently H, C_1-C_6 alkyl or C_3-C_8 cycloalkyl; or R^5 and R^6, or R^7 and R^8 may be joined together to form a 3 to 8 membered carbocyclic ring;

X is a 4-10 membered mono or bicyclic heterocyclic

group containing carbon ring atoms and one ring nitrogen atom through which the group is attached to the adjacent carbonyl group; the group may be saturated or partially unsaturated and, in addition to the -$(CR^7R^8)_m$-Het substituent, it may be substituted by up to 4 further substituents each independently chosen from F, C_1-C_6 alkyl, C_3-C_8 cycloalkyl, OR^{11} or NR^9R^{10};

Het is an imidazolyl or triazolyl group either of which may optionally be substituted by C_1-C_6 alkyl, C_3-C_8 cycloalkyl, NR^9R^{10} or $CONR^9R^{10}$,

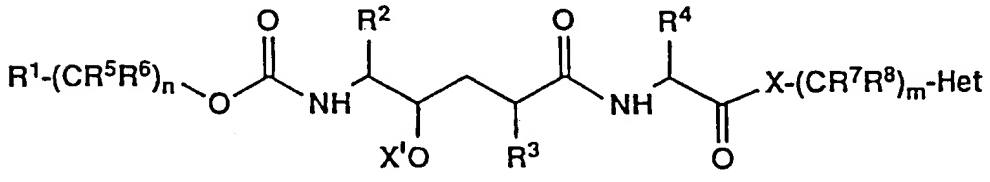
each of R^9 and R^{10} is independently H, C_1-C_6 alkyl or C_3-C_8 cycloalkyl, or R^9 and R^{10} may be joined together to form, with the nitrogen to which they are attached, a 4 to 8 membered nitrogen-containing heterocyclic group,

R^{11} is H, C_1-C_6 alkyl or C_3-C_8 cycloalkyl;

n and m are each independently 0, 1 or 2;

wherein any alkyl or cycloalkyl group included in the aforementioned definitions may optionally be fully or partially substituted by fluorine;

which comprises removing the protecting group from a compound of the formula:

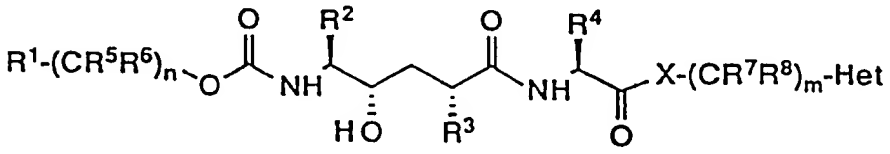


(IX)

wherein X^1 is a selectively removable hydroxy-protecting group and R^1 to R^8, X and Het are as defined in claim 1, and isolating the compound of formula (I) and optionally forming a pharmaceutically acceptable salt thereof.

16. A process as claimed in claim 1 wherein the selectively removable hydroxy protecting group X^1 is t-butyldimethylsilyl and it is removed by reaction with tetra-n-butylammonium floride in an organic solvent.

17. A process as claimed in claim 1 wherein the compound of formula (I) has the stereochemistry-:



wherein R^1 to R^8, n, m, X and Het are as previously defined in claim 1.

18. A process as claimed in claim 1, claim 2 or claim 3 wherein R^1 is t-butyl, isopropyl, or oxetanyl and n is 0.

19. A process as claimed in claim 4 wherein R^2 is benzyl.

20. A process as claimed in claim 4 or 5 wherein R^4 is isopropyl or sec-butyl.

21. A process is claimed in any one of claims 4 to 6 wherein R^3 is benzyl optionally substituted in the

phenyl ring by methyl, fluoro, chloro, iodo, CF_3, or OCF_3, or R^3 is 3-phenylpropyl or 3-phenyl-prop-2-enyl.

22. A process as claimed in any one of claims 4 to 7 wherein each of R^5, R^6, R^7 and R^8 is H.

23. A process as claimed in any one of claims 4 to 8 wherein m is 0 or 1.

24. A process as claimed in claim 1 wherein said compound of formula I produced is-:

1-[N-((R)-2-benzyl-(S)-5-(t-butoxycarbonylamino)-(S)-4-hydroxy-6-phenylhexanoyl)-(S)-valyl]-3-(imidazol-1-yl)azetidine,

1-[N-((R)-2-benzyl-(S)-5-(t-butoxycarbonylamino)-(S)-4-hydroxy-6-phenylhexanoyl)-(S)-valyl]-4-(imidazol-1-yl)piperidine,

1-[N-((S)-5-(t-butoxycarbonylamino)-(S)-4-hydroxy-6-phenyl-(R)-2-(3-phenylprop-2-en-1-yl)hexanoyl)-(S)-valyl]-4-(imidazol-1-yl)piperidine,

1-[N-((S)-5-(t-butoxycarbonylamino)-(S)-4-hydroxy-6-phenyl-(R)-2-(4-trifluoromethoxybenzyl)hexanoyl)-(S)-valyl]-4-(imidazol-1-yl)piperidine,

1-[N-((S)-5-(t-butoxycarbonylamino)-(R)-2-(4-chlorobenzyl)-(S)-4-hydroxy-6-phenylhexanoyl)-(S)-valyl]-3-(imidazol-1-yl)azetidine or

1-[N-((S)-5-(t-butoxycarbonylamino)-(S)-4-hydroxy-6-phenyl-(R)-2-(4-trifluoromethoxybenzyl)hexanoyl)-(S)-isoleucyl]-4-(imidazol-1-yl)piperidine.

INTERNATIONAL SEARCH REPORT

International Application No PCT/EP 93/00597

I. CLASSIFICATION OF SUBJECT MATTER (if several classification symbols apply, indicate all)[6]

According to International Patent Classification (IPC) or to both National Classification and IPC

Int.Cl. 5 C07D403/04; C07D401/04; A61K31/41; A61K31/445

II. FIELDS SEARCHED

Minimum Documentation Searched[7]

Classification System	Classification Symbols
Int.Cl. 5	C07D ; A61K ; C07K

Documentation Searched other than Minimum Documentation to the Extent that such Documents are Included in the Fields Searched[8]

III. DOCUMENTS CONSIDERED TO BE RELEVANT[9]

Category °	Citation of Document, [11] with indication, where appropriate, of the relevant passages [12]	Relevant to Claim No.[13]
A	EP,A,0 386 611 (F. HOFFMANN-LA ROCHE AG) 12 September 1990 see claims	1-24
A	EP,A,0 437 729 (BAYER AG) 24 July 1991 see claims	1-24
A	EP,A,0 365 992 (ABBOTT LABORATORIES) 2 May 1990 see claims	1-24
A	WO,A,9 012 804 (THE UPJOHN COMPANY) 1 November 1990 see claims	1-24
A	WO,A,9 003 971 (ABBOTT LABORATORIES) 19 April 1990 see claims	1-24

° Special categories of cited documents : [10]

"A" document defining the general state of the art which is not considered to be of particular relevance

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"P" document published prior to the international filing date but later than the priority date claimed

"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step

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"&" document member of the same patent family

IV. CERTIFICATION

Date of the Actual Completion of the International Search	Date of Mailing of this International Search Report
18 JUNE 1993	25. 06. 93
International Searching Authority	**Signature of Authorized Officer**
EUROPEAN PATENT OFFICE	CHOULY J.

Form PCT/ISA/210 (second sheet) (January 1985)

ANNEX TO THE INTERNATIONAL SEARCH REPORT ON INTERNATIONAL PATENT APPLICATION NO.

EP 9300597
SA 71545

This annex lists the patent family members relating to the patent documents cited in the above-mentioned international search report. The members are as contained in the European Patent Office EDP file on
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